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(54) Title: DNA SEQUENCES AND PLASMIDS FOR THE PREPARATION OF SUGAR BEET WITH CHANGED SUCROSE CONCENTRATION (57) Abstract DNA sequences and plasmids are described, that by integration in a plant genome of sugar beet change the sucrose concentration, as well as transgenic plants that by introduction of the DNA sequences of the invention causes changes in sugar concentration.		

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Title: DNA sequences and plasmids for the preparation of sugar beet with changed sucrose concentration

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Field of the invention

The present invention relates to DNA sequences and plasmids, containing these DNA sequences, which by integration into the genome of a sugar-beet plant, changes
10 the sugar metabolism of the plant to be changed. The invention also relates to transgenic plants formed with the help of these sequences.

Sucrose is of central importance for the plant and serves
15 many functions. For the long distance transport of photoassimilates and/or energy between various organs in plants, sucrose is almost exclusively used. The sucrose, which is transported in a specific heterotrophic organ, determines the growth and the development of this organ.
20 Thus it is known, e.g. from EP 442 592, that transgenic plants, in which the transport away of the sucrose from the exporting leaves is inhibited by expression of an apoplastic invertase, shows a strong reduction in the growth of e.g. roots or tubers in the case of potato
25 plants. For tobacco plants, the principal importance of sucrose as the central function for the long distance transport of energy carriers within the plant is described (von Schaewen et al, 1990, EMBO J 9: 3033-3044).

30 Further it is also known from EP 455 316 that DNA sequences present on plasmids, after introduction in a plant genome of a potato plant can affect the starch biosynthesis as well altering the amount and composition of the protein in the potato tubers.

35

Whilst it has known that a reduction of the amount of sucrose imported in the heterotrophic organs, such as tubers and seeds, leads to loss of yield, it is not known whether an increase in the amount of sucrose in the photosynthetically active parts of the plant, mainly the leaves, leads to a better supply of heterotrophic organs and thus to an increase in yield.

Besides sucrose and/or the hexoses, glucose and fructose, derived from sucrose, have the property of protection of plants against frost damage at low temperatures. Frost damage is one of the main limiting factors in agricultural productivity in the northern hemisphere. Temperatures below freezing lead to the formation of ice crystals. Since the growing ice crystals consist of pure water, water is abstracted from the cells as the temperature falls.

This dehydration has at least two potential damaging results:

1. All dissolved substances within a cell are strongly concentrated and the cell contracts following the loss of water. Highly concentrated salts and organic acids lead to membrane damage.

2. With rehydration from dew, the previously contacted cells reexpand. The cell membrane also expands again. The volume expansion puts a heavy mechanical load on the membrane.

It is thus clear that a freezing/dew cycle can lead to severe membrane damage of the cells and thus to damage to the plant.

It thus appears worth trying to hinder the freezing. One

possible strategy is the increased formation of osmotically active substances in the cytosol of plant cells. This should lead to a lowering of the freezing point. Osmotically active substances include sucrose
5 and/or the two hexoses derived from sucrose.

The increased formation of sucrose and/or the two hexoses at low temperatures is desirable in the growing plant. Another situation can exist in the harvested parts of a
10 plant, especially in storage.

In relation to the economic aspects, sucrose thus possesses two especially important functions:

- 15 1 as the transport form for the distant transport of photoassimilates,
- 2 as an osmotically active substance with the desirable activity of lowering the freezing point in intact, growing plants.

20 The biosynthesis pathways for the formation of sucrose, either from the primary photosynthesis products (in the leaf) or by breakdown of starch (in the storage organs e.g. of potatoes), are known.

25 It is however, not known how and in what way changes of the carbohydrate concentration in sugar beet can be achieved since it is not possible to use even very similar genes such as for example genes that code for a sucrose synthase, ADP-glucose pyrophosphorylase or sucrose
30 phosphate synthase of the potato with satisfactory success for the preparation of sugar beet with changed sucrose concentration. An exact analysis and determination of the DNA sequences or sequence fragments for the sugar beet is thus required.

35

To change the sugar concentration in sugar beet, DNA sequences are now provided which code for the small and large subunit of the ADP glucose pyrophosphorylase, the sucrose synthase and the sucrose phosphate synthase of sugar beet (Seq. ID No 1-4).

These DNA sequences can be introduced into plasmids and in this way combined with steering elements for expression in eukaryotic cells. Such steering elements are on the one hand transcription promoters and on the other hand transcription terminators. Each plasmid comprises:

- a) a suitable promoter which ensures that the coding sequence meets a suitable time point or in the specified developments in the transgenic plant or in determined genes of transgenic plants;
- b) at least a coding sequence for sugar beet that
 - i) is so coupled to the promoter that the formation of an RNA is allowed which is into a protein, whereby the protein demonstrates an enzymatic activity which leads to a change of the sucrose concentration in the plant, or
 - ii) which is so coupled to the promoter that the coding part is read, which leads to the formation of a so-called anti-sense RNA which under-expresses the formation of the protein coded from an endogenous gene in the plant, that is involved in the sucrose biosynthesis; and
- c) A non-coding termination sequence that contains the signal for the termination and polyadenylation of the transcript.

5

The coding sequences named under b) are the sequences that code for the large and small subunit of the ADP glucose pyrophosphorylase, for the sucrose phosphate synthase and for the sucrose synthase of sugar beet.

5

The large subunit of the ADP-glucose-pyrophosphorylase has the following nucleotide sequence (Seq. ID No. 1):

CAAAAGAAAA	ACTTCCCATT	TCTACTTCTT	TGCACAATAT	AATTTCCCAC	0050										
CAATTTTCT	TTAAATTTCT	CACTTTCATT	TAATCAGTTT	TCAGCAACAT	0100										
TCTGATACTC	GACAACCCAC	TTTCTGTTCT	CCCAAGATTC	CAAACCTCTG	0150										
ATTCTCATTC	CACTAATATT	TTTGCTTATT	TTTTTTCTGG	ATTAAAGAA	0200										
AAGCT	ATG	GAT	GCA	AGT	GCA	GCA	GCC	ATA	AAT	GTC	AAT	GCC	CAT	0243	
	Met	Asp	Ala	Ser	Ala	Ala	Ala	Ile	Asn	Val	Asn	Ala	His		
					5								10		
TTA	ACA	GAA	GTT	GGA	AAG	AAA	CGT	TTT	TTA	GGA	GAG	AGA	ATC	AGT	0288
Leu	Thr	Glu	Val	Gly	Lys	Lys	Arg	Phe	Leu	Gly	Glu	Arg	Ile	Ser	
	15					20								25	
CAA	AGT	TTG	AAG	GGT	AAA	GAT	CTG	AGA	GCT	CTG	TTT	TCA	AGA	ACT	0333
Gln	Ser	Leu	Lys	Gly	Lys	Asp	Leu	Arg	Ala	Leu	Phe	Ser	Arg	Thr	
	30					35								40	
GAG	AGC	AAG	GGT	AGA	AAT	GTC	AAT	AAA	CCT	GGG	GTT	GCA	TTT	TCT	0378
Glu	Ser	Lys	Gly	Arg	Asn	Val	Asn	Lys	Pro	Gly	Val	Ala	Phe	Ser	
	45					50								55	
GTT	CTC	ACC	TCA	GAT	TTT	AAT	CAA	AGT	GTT	AAA	GAA	TCT	TTG	AAA	0423
Val	Leu	Thr	Ser	Asp	Phe	Asn	Gln	Ser	Val	Lys	Glu	Ser	Leu	Lys	
	60					65								70	

6

TAT	GAG	CCA	GCA	TTA	TTT	GAA	TCT	CCA	AAA	GCT	GAC	CCA	AAA	AAT	0468
Tyr	Glu	Pro	Ala	Leu	Phe	Glu	Ser	Pro	Lys	Ala	Asp	Pro	Lys	Asn	
75						80					85				
GTG	GCT	GCA	ATT	GTG	CTG	GGT	GGT	GGT	GCT	GGG	ACT	CGC	CTC	TTT	0513
Val	Ala	Ala	Ile	Val	Leu	Gly	Gly	Gly	Ala	Gly	Thr	Arg	Leu	Phe	
90						95					100				
CCT	CTT	ACT	AGC	AGG	AGA	GCT	AAG	CCA	GCA	GTG	CCA	ATT	GGA	GGG	0558
Pro	Leu	Thr	Ser	Arg	Arg	Ala	Lys	Pro	Ala	Val	Pro	Ile	Gly	Gly	
105						110					115				
TGT	TAC	AGG	CTG	ATT	GAT	GTG	CCT	ATG	AGC	AAC	TGC	ATC	AAC	AGT	0603
Cys	Tyr	Arg	Leu	Ile	Asp	Val	Pro	Met	Ser	Asn	Cys	Ile	Asn	Ser	
120						125					130				
GGC	ATT	AGA	AAG	ATT	TTC	ATT	CTT	ACC	CAG	TTC	AAT	TCG	TTT	TCG	0648
Gly	Ile	Arg	Lys	Ile	Phe	Ile	Leu	Thr	Gln	Phe	Asn	Ser	Phe	Ser	
135						140					145				
CTT	AAT	CGT	CAT	CTT	GCT	CGA	ACC	TAT	AAT	TTT	GGA	GAT	GGT	GTG	0693
Leu	Asn	Arg	His	Leu	Ala	Arg	Thr	Tyr	Asn	Phe	Gly	Asp	Gly	Val	
150						155					160				
AAT	TTT	GGG	GAT	GGC	TTT	GTG	GAG	GTT	TTT	GCT	GCT	ACA	CAA	ACA	0738
Asn	Phe	Gly	Asp	Gly	Phe	Val	Glu	Val	Phe	Ala	Ala	Thr	Gln	Thr	
165						170					175				

7

CCT GGA GAA TCA GGA AAG AAA TGG TTC CAG GGC ACC GCT GAT GCA 0783
 Pro Gly Glu Ser Gly Lys Lys Trp Phe Gln Gly Thr Ala Asp Ala
 180 185 190

GTA AGA CAG TTT TTC TGG GCA TTT GAG GAT TCC AAA TCC AAG GAT 0828
 Val Arg Gln Phe Phe Trp Ala Phe Glu Asp Ser Lys Ser Lys Asp
 195 200 205

GTC GAG CAT ATA GTT ATT TTA TCC GGT GAT CAT CTT TAC CGA ATG 0873
 Val Glu His Ile Val Ile Leu Ser Gly Asp His Leu Tyr Arg Met
 210 215 220

GAT TAC ATG AGT TTT TGG CAG AAG CAC ATT GAC ACC AAT GCT GAT 0918
 Asp Tyr Met Ser Phe Trp Gln Lys His Ile Asp Thr Asn Ala Asp
 225 230 235

ATT ACA GTG TCA TGC ATA CCC ATG GAT GAC AGC CGT GCA TCG GAT 0963
 Ile Thr Val Ser Cys Ile Pro Met Asp Asp Ser Arg Ala Ser Asp
 240 245 250

TAT GGG CTG ATG AAG ATT GAT CAC ACT GGA CGC ATT GTC CAT TTT 1008
 Tyr Gly Leu Met Lys Ile Asp His Thr Gly Arg Ile Val His Phe
 255 260 265

GCA GAA AAA CCC AAG GGT TCT GAT CTA ACA GCA ATG CAA GTA GAT 1053
 Ala Glu Lys Pro Lys Gly Ser Asp Leu Thr Ala Met Gln Val Asp
 270 275 280

8

ACA ACT GTT CTT GGG CTC TCT GAC CTT GAA GCT ATG TCA AAT CCA 1098
 Thr Thr Val Leu Gly Leu Ser Asp Leu Glu Ala Met Ser Asn Pro
 285 290 295

TAT ATT GCA TCA ATG GGT GTT TAT GTC TTT CGA ACG GAT GTT CTT 1143
 Tyr Ile Ala Ser Met Gly Val Tyr Val Phe Arg Thr Asp Val Leu
 300 305 310

ATG GAG CTT CTC AAT CGA AAA TAC CCT TCA AGC AAT GAT TTT GGC 1188
 Met Glu Leu Leu Asn Arg Lys Tyr Pro Ser Ser Asn Asp Phe Gly
 315 320 325

TCT GAG ATT ATT CCT TCA GCT GTA GGA GAG TCT AAT GTT CAG GCA 1233
 Ser Glu Ile Ile Pro Ser Ala Val Gly Glu Ser Asn Val Gln Ala
 330 335 340

TAT CTA TTT AAT GAC TAC TGG GAG GAT ATC GGA ACC ATA AAG TCT 1278
 Tyr Leu Phe Asn Asp Tyr Trp Glu Asp Ile Gly Thr Ile Lys Ser
 345 350 355

TTC TTT GAT TCC AAT TTG GCC CTT ACA CAA CAG CCT CCC AAG TTT 1323
 Phe Phe Asp Ser Asn Leu Ala Leu Thr Gln Gln Pro Pro Lys Phe
 360 365 370

GAA TTC TAC GAT CCA AAA ACA CCT TTT TAT ACA TCT GCA AGA TTT 1368
 Glu Phe Tyr Asp Pro Lys Thr Pro Phe Tyr Thr Ser Ala Arg Phe
 375 380 385

9

CTG CCT CCT ACA AAA GTC GAC AGG TGC AAG ATT GTC GAT TCC ATT 1413
 Leu Pro Pro Thr Lys Val Asp Arg Cys Lys Ile Val Asp Ser Ile
 390 395 400

GTA TCC CAT GGT TGT TTT CTA CAG GAG TCT AGC ATC CAA CAT TCC 1458
 Val Ser His Gly Cys Phe Leu Gln Glu Ser Ser Ile Gln His Ser
 405 410 415

ATT GTT GGT GTT CGC TCA AGA TTA GAG TCC GGG GTT GAG TTC CAG 1503
 Ile Val Gly Val Arg Ser Arg Leu Glu Ser Gly Val Glu Phe Gln
 420 425 430

GAC ACC ATG ATG ATG GGC GCA GAT TAC TAT CAA ACT GAA TCA GAA 1548
 Asp Thr Met Met Met Gly Ala Asp Tyr Tyr Gln Thr Glu Ser Glu
 435 440 445

ATT GCT TCT CTG CTT GCT GAG GGA AAG GTT CCT GTT GGT GTC GGA 1593
 Ile Ala Ser Leu Leu Ala Glu Gly Lys Val Pro Val Gly Val Gly
 450 455 460

CAG AAT ACC AAA ATA AAG AAT TGC ATA ATT GAC AAG AAC GCC AAA 1638
 Gln Asn Thr Lys Ile Lys Asn Cys Ile Ile Asp Lys Asn Ala Lys
 465 470 475

ATT GGA AAA GAT GTG GTA ATC GCA AAC ACG GAT GGT GTT GAG GAA 1683
 Ile Gly Lys Asp Val Val Ile Ala Asn Thr Asp Gly Val Glu Glu
 480 485 490

10

GCA GAT AGA CCA AAT GAA GGC TTT TAC ATC AGG TCG GGC ATT ACC 1728
 Ala Asp Arg Pro Asn Glu Gly Phe Tyr Ile Arg Ser Gly Ile Thr
 495 500 505

ATC ATT TTG AAG AAC GCA ACC ATA CAA GAC GGT CTT GTG ATT TAG 1773
 Ile Ile Leu Lys Asn Ala Thr Ile Gln Asp Gly Leu Val Ile End
 510 515 520

ATTTAATCAT AACCTCATTA GAAAGAAATA ATTTTGCATG ATTCCTTTT 1823

CATGTAACCT AAAGTGGCTA AACCACGAGG TTTTCTCATC TGTATATATA 1873

ATATGTCTAT AACTATGGAT AATCTTAATA AAAAAAAAAA AAAAAAAAAA 1923

A 1924

The small subunit of the ADP-glucose-pyrophosphatase has
 the following nucleotide sequence (Seq. ID No. 2):

GG ATA ACT GTG CCA TCA ACC TCC TCA AAG AAC CTC CAA AAT AGC 0044
 Ile Thr Val Pro Ser Thr Ser Ser Lys Asn Leu Gln Asn Ser
 5 10

CTC GCA TTC TCC TCT TCT TCT CTC TCC GGC GAC AAA ATT CAA ACG 0089
 Leu Ala Phe Ser Ser Ser Ser Leu Ser Gly Asp Lys Ile Gln Thr
 15 20 25

ACG TCA TTT CTC AAC CGC CGA TAT TGT AGA ATC TCT TCT AGA GCT 0134
 Thr Ser Phe Leu Asn Arg Arg Tyr Cys Arg Ile Ser Ser Arg Ala
 30 35 40

11

CCG ATT GTT GTC TCT CCC AAA GCT GTT TCT GAT TCT AAG AAT TCG	0179
Pro Ile Val Val Ser Pro Lys Ala Val Ser Asp Ser Lys Asn Ser	
45 50 55	
CAG ACT TGT CTT GAC CCT GAA GCC AGC CGT AGT GTT CTT GGT ATT	0224
Gln Thr Cys Leu Asp Pro Glu Ala Ser Arg Ser Val Leu Gly Ile	
60 65 70	
ATA CTT GGA GGT GGT GCT GGT ACA CGT CTT TAC CCG TTG ACT AAG	0269
Ile Leu Gly Gly Gly Ala Gly Thr Arg Leu Tyr Pro Leu Thr Lys	
75 80 85	
AAG AGA GCC AAA CCA GCC GTG CCA CTC GGT GCT AAT TAT AGG CTT	0314
Lys Arg Ala Lys Pro Ala Val Pro Leu Gly Ala Asn Tyr Arg Leu	
90 95 100	
ATT GAT ATC CCA GTG AGC AAT TGT TTG AAC AGT AAT ATT TCC AAA	0359
Ile Asp Ile Pro Val Ser Asn Cys Leu Asn Ser Asn Ile Ser Lys	
105 110 110	
ATA TAT GTT CTT ACA CAA TTC AAT TCT GCT TCT CTG AAT CGT CAT	0404
Ile Tyr Val Leu Thr Gln Phe Asn Ser Ala Ser Leu Asn Arg His	
115 120 125	
CTT TCG CGG GCA TAT GCT AGC AAC ATG GGA GGA TAC AAA AAT GAG	0449
Leu Ser Arg Ala Tyr Ala Ser Asn Met Gly Gly Tyr Lys Asn Glu	
130 135 140	
GGG TTT GTA GAA GTT CTT GCT GCT CAG CAA AGT CCA GAG AAT CCA	0494
Gly Phe Val Glu Val Leu Ala Ala Gln Gln Ser Pro Glu Asn Pro	
145 150 155	

12

AAC TGG TTT CAG GGT ACA GCT GAT GCT GTT AGG CAA TAT CTG TGG 0539
 Asn Trp Phe Gln Gly Thr Ala Asp Ala Val Arg Gln Tyr Leu Trp
 160 165 170

CTT TTC GAA GAG CAC AAT GTT CTT GAG TAC TTG ATT CTT GCT GGT 0584
 Leu Phe Glu Glu His Asn Val Leu Glu Tyr Leu Ile Leu Ala Gly
 175 180 185

GAC CAT TTG TAT CGA ATG GAT TAT GAA AGA TTT GTC CAA GCT CAC 0629
 Asp His Leu Tyr Arg Met Asp Tyr Glu Arg Phe Val Gln Ala His
 190 195 200

AGA GAA ACT GAT GCA GAC ATT ACT GTT GCT GCA TTG CCA ATG GAT 0674
 Arg Glu Thr Asp Ala Asp Ile Thr Val Ala Ala Leu Pro Met Asp
 205 210 215

GAA AAG CGT GCT ACT GCA TTT GGT TTG ATG AAA ATT GAT GAA GAA 0719
 Glu Lys Arg Ala Thr Ala Phe Gly Leu Met Lys Ile Asp Glu Glu
 220 225 230

GGA AGA ATT ATT GAG TTT GCC GAG AAA CCG AAA GGA GAA CAA TTG 0764
 Gly Arg Ile Ile Glu Phe Ala Glu Lys Pro Lys Gly Glu Gln Leu
 235 240 245

AAA GCT ATG AAG GTT GAT ACC ACA ATC CTG GGT CTG GAC GAT GAG 0809
 Lys Ala Met Lys Val Asp Thr Thr Ile Leu Gly Leu Asp Asp Glu
 250 255 260

13

AGA	GCA	AAA	GAA	ATG	CCA	TTC	ATA	GCC	AGC	ATG	GGC	ATA	TAT	GTT	0854
Arg	Ala	Lys	Glu	Met	Pro	Phe	Ile	Ala	Ser	Met	Gly	Ile	Tyr	Val	
265					270					275					

ATT	AGC	AAA	GAT	GTA	ATG	CTT	AAT	CTG	CTT	CGG	GAG	CAA	TTT	CCT	0899
Ile	Ser	Lys	Asp	Val	Met	Leu	Asn	Leu	Leu	Arg	Glu	Gln	Phe	Pro	
280					285					290					

GGT	GCT	AAT	GAT	TTT	GGA	AGT	GAA	GTT	ATT	CCA	GGC	GCC	ACT	TCC	0944
Gly	Ala	Asn	Asp	Phe	Gly	Ser	Glu	Val	Ile	Pro	Gly	Ala	Thr	Ser	
295					300					305					

ATA	GGG	TTG	AGA	GTC	CAA	GCT	TAT	TTG	TAT	GAT	GGT	TAC	TGG	GAG	0989
Ile	Gly	Leu	Arg	Val	Gln	Ala	Tyr	Leu	Tyr	Asp	Gly	Tyr	Trp	Glu	
310					315					320					

GAT	ATT	GGT	ACC	ATT	GAA	GCT	TTT	TAC	AAT	GCT	AAC	TTG	GGA	ATC	1034
Asp	Ile	Gly	Thr	Ile	Glu	Ala	Phe	Tyr	Asn	Ala	Asn	Leu	Gly	Ile	
325					330					335					

ACC	AAA	AAG	CCG	GTG	CCA	GAT	TTT	AGC	TTC	TAT	GAT	CGT	TCA	TCT	1079
Thr	Lys	Lys	Pro	Val	Pro	Asp	Phe	Ser	Phe	Tyr	Asp	Arg	Ser	Ser	
340					345					350					

CCA	ATT	TAT	ACA	CAA	CCT	CGG	TAT	TTG	CCT	CCT	TCA	AAG	ATG	CTT	1124
Pro	Ile	Tyr	Thr	Gln	Pro	Arg	Tyr	Leu	Pro	Pro	Ser	Lys	Met	Leu	
355					360					365					

14

GAT GCT GAT ATA ACT GAC AGC GTC ATC GGT GAA GGC TGT GTT ATT 1169
Asp Ala Asp Ile Thr Asp Ser Val Ile Gly Glu Gly Cys Val Ile
370 375 380

AAG AAC TGT AAG ATT CAT CAT TCT GTT ATC GGA CTT CGA TCT TGT 1214
Lys Asn Cys Lys Ile His His Ser Val Ile Gly Leu Arg Ser Cys
385 390 395

ATC TCG GAG GGT GCA ATC ATT GAG GAC ACA CTG TTG ATG GGA GCT 1259
Ile Ser Glu Gly Ala Ile Ile Glu Asp Thr Leu Leu Met Gly Ala
400 405 410

GAT TAT TAT GAG ACT GAT GCT GAT CGG AAA TTC CTG GCT GCT AAG 1304
Asp Tyr Tyr Glu Thr Asp Ala Asp Arg Lys Phe Leu Ala Ala Lys
415 420 425

GGT AGT GTA CCT ATT GGA ATT GGG AAT GCA CGT ATT GGG GAT GAT 1349
Gly Ser Val Pro Ile Gly Ile Gly Asn Ala Arg Ile Gly Asp Asp
430 435 440

GTC AAG ATT ATC AAC AGT GAC AAT GTA CAA GAA GCA GCA AGA GAA 1394
Val Lys Ile Ile Asn Ser Asp Asn Val Gln Glu Ala Ala Arg Glu
445 450 455

ACA GAC GGA TAC TTC ATA AAG AGC GGA ATA GTC ACT ATA ATC AAG 1439
Thr Asp Gly Tyr Phe Ile Lys Ser Gly Ile Val Thr Ile Ile Lys
460 465 470

15

GAC GCC ATG ATT CCA AGT GGA ACT GTA ATC TAG AAATGGAGCA 1482
 Asp Ala Met Ile Pro Ser Gly Thr Val Ile End
 475 480 485

TATAATAAAT ATCACTGCCT ATTTACAGTA CCTATCTGAG TCTCCCACCA 1532

TGACCCTTTG ATTCAATCTT TTAGTTATGT AAATATTTTT GGCTTTTGCG 1582

ATTTTGCCAT AAATTGAAG AAGCGAGGAT TCAGGGACGA TAGTGCTATG 1632

AATTGGAAGA AAGGATTTGG GGGATATCTT TGTAAGACA TTTTGACTAC 1682

TGGGCACTAA AAATTGCGTA ATGCTATACC AAAATATATA AAAAGATCTT 1732
 GCTGGGTTTT GTAAAAAAA AAAAAAAAAA A 1763

The sucrose phosphate - synthase has the following
 nucleotide sequence (Seq. ID No. 3):

GGGCTGCAGG GAAGCTCTGA ACTTCAAAA ATG GCG GGA AAT GAT 0044
 Met Ala Gly Asn Asp
 5

TGG ATA AAC AGT TAT TTA GAG GCA ATT CTG GAT GTG GGT CCA GGA 0089
 Trp Ile Asn Ser Tyr Leu Glu Ala Ile Leu Asp Val Gly Pro Gly
 10 15 20

CTT GAT GAT GCA AAA TCA TCT TTG CTT TTG AGA GAA AGA GGC AGG 0134
 Leu Asp Asp Ala Lys Ser Ser Leu Leu Leu Arg Glu Arg Gly Arg
 25 30 35

TTT AGT CCT ACT CGT TAC TTT GTT GAA GAA GTT ATC ACT GGT TTT 0179
 Phe Ser Pro Thr Arg Tyr Phe Val Glu Glu Val Ile Thr Gly Phe
 40 45 50

16

GAT	GAA	ACC	GAC	CTT	CAT	CGT	TCA	TGG	GTT	CGG	GCA	CAA	GCA	ACA	0224
Asp	Glu	Thr	Asp	Leu	His	Arg	Ser	Trp	Val	Arg	Ala	Gln	Ala	Thr	
				55					60					65	

AGG	AGT	CCT	CAA	GAG	AGG	AAT	ACT	AGA	TTG	GAG	AAC	ATG	TGT	TGG	0269
Arg	Ser	Pro	Gln	Glu	Arg	Asn	Thr	Arg	Leu	Glu	Asn	Met	Cys	Trp	
				70					75					80	

AGA	ATT	TGG	AAT	TTG	GCT	CGT	CAG	AAG	AAG	CAG	CTT	GAG	AAT	GAA	0314
Arg	Ile	Trp	Asn	Leu	Ala	Arg	Gln	Lys	Lys	Gln	Leu	Glu	Asn	Glu	
				85					90					95	

GAA	GCT	CAG	CGG	AAG	ACA	AAA	CGT	CGT	ATG	GAG	CTT	GAG	AGG	GGT	0359
Glu	Ala	Gln	Arg	Lys	Thr	Lys	Arg	Arg	Met	Glu	Leu	Glu	Arg	Gly	
				100					105					110	

CGT	CGA	GAA	GCA	ACT	GCT	GAT	ATG	TCG	GAG	GAC	TTA	TCA	GAA	GGC	0404
Arg	Arg	Glu	Ala	Thr	Ala	Asp	Met	Ser	Glu	Asp	Leu	Ser	Glu	Gly	
				115					120					125	

GAA	AAG	GAC	ATT	TCA	GCT	CAT	GGT	GAT	AGC	ACC	CGT	CCT	AGA	TTG	0449
Glu	Lys	Asp	Ile	Ser	Ala	His	Gly	Asp	Ser	Thr	Arg	Pro	Arg	Leu	
				130					135					140	

CCA	AGA	ATA	AAT	TCT	CTT	GAT	GCT	ATG	GAG	ACA	TGG	ATT	AGT	CAA	0494
Pro	Arg	Ile	Asn	Ser	Leu	Asp	Ala	Met	Glu	Thr	Trp	Ile	Ser	Gln	
				145					150					155	

CAA	AAG	GAA	AAA	AAA	CTC	TAC	CTT	GTT	TTG	ATA	AGT	CTT	CAT	GGT	0539
Gln	Lys	Glu	Lys	Lys	Leu	Tyr	Leu	Val	Leu	Ile	Ser	Leu	His	Gly	
				160					165					170	

17

TTG ATA CGA GGT GAA AAC ATG GAA CTT GGC CGT GAT TCT GAT ACT 0584
Leu Ile Arg Gly Glu Asn Met Glu Leu Gly Arg Asp Ser Asp Thr
175 180 185

GGT GGT CAG GTT AAG TAT GTG GTT GAG CTT GCA AGG GCT CTA GGT 0629
Gly Gly Gln Val Lys Tyr Val Val Glu Leu Ala Arg Ala Leu Gly
190 195 200

TCG ATG CCA GGT GTT TAT AGA GTT GAT TTG CTA ACT AGG CAA GTT 0674
Ser Met Pro Gly Val Tyr Arg Val Asp Leu Leu Thr Arg Gln Val
205 210 215

TCA TCT CCT GAC GTG GAT TGG AGT TAT GGG GAG CCT ACT GAG ATG 0719
Ser Ser Pro Asp Val Asp Trp Ser Tyr Gly Glu Pro Thr Glu Met
220 225 230

CTG AAT CCA AGG GAT TCC AAT GGT TTT GAT GAT GAT GAT GAT GAA 0764
Leu Asn Pro Arg Asp Ser Asn Gly Phe Asp Asp Asp Asp Asp Glu
235 240 245

ATG GGA GAG AGT AGT GGT GCT TAC ATT GTT CGT ATA CCA TTT GGG 0809
Met Gly Glu Ser Ser Gly Ala Tyr Ile Val Arg Ile Pro Phe Gly
250 255 260

CCG AGG GAT AAG TAT ATC GCA AAA GAA GAG CTT TGG CCC TAT ATT 0854
Pro Arg Asp Lys Tyr Ile Ala Lys Glu Glu Leu Trp Pro Tyr Ile
265 270 275

18

CCT	GAA	TTT	GTT	GAT	GGT	GCT	CTA	AAC	CAC	ATA	GTT	CAA	ATG	TCC	0899
Pro	Glu	Phe	Val	Asp	Gly	Ala	Leu	Asn	His	Ile	Val	Gln	Met	Ser	
				280				285					290		

AAA	GTT	TTA	GGT	GAG	CAA	ATT	GGT	AGC	GGG	GAA	ACA	GTT	TGG	CCA	0944
Lys	Val	Leu	Gly	Glu	Gln	Ile	Gly	Ser	Gly	Glu	Thr	Val	Trp	Pro	
				295				300					305		

GTT	GCC	ATT	CAT	GGA	CAT	TAT	GCT	GAT	GCT	GGT	GAT	TCT	GCT	GCT	0989
Val	Ala	Ile	His	Gly	His	Tyr	Ala	Asp	Ala	Gly	Asp	Ser	Ala	Ala	
				310				315					320		

CTT	CTT	TCT	GGT	GGC	CTA	AAT	GTT	CCA	ATG	CTT	TTA	ACG	GGG	CAT	1034
Leu	Leu	Ser	Gly	Gly	Leu	Asn	Val	Pro	Met	Leu	Leu	Thr	Gly	His	
				325				330					335		

TCT	CTT	GGC	CGA	GAC	AAG	TTA	GAG	CAG	CTC	CTC	AAA	CAG	GGT	CGA	1079
Ser	Leu	Gly	Arg	Asp	Lys	Leu	Glu	Gln	Leu	Leu	Lys	Gln	Gly	Arg	
				340				345					350		

ATG	TCT	AAA	GAT	GAC	ATA	AAC	AAT	ACA	TAC	AAA	ATA	ATG	CGT	AGG	1124
Met	Ser	Lys	Asp	Asp	Ile	Asn	Asn	Thr	Tyr	Lys	Ile	Met	Arg	Arg	
				355				360					365		

ATA	GAA	GCC	GAA	GAG	TTA	TCA	CTT	GAT	GCC	TCT	GAG	ATA	GTC	ATA	1169
Ile	Glu	Ala	Glu	Glu	Leu	Ser	Leu	Asp	Ala	Ser	Glu	Ile	Val	Ile	
				370				375					380		

19

ACT AGT ACA AGA CAA GAA ATA GAA GAG CAA TGG CAC CTC TAT GAT 1214
 Thr Ser Thr Arg Gln Glu Ile Glu Glu Gln Trp His Leu Tyr Asp
 385 390 395

GGG TTT GAT CCT GTG CTA GAA CGT AAA CTC CGT GCT AGG ATG AAG 1259
 Gly Phe Asp Pro Val Leu Glu Arg Lys Leu Arg Ala Arg Met Lys
 400 405 410

CGT GGT GTA AGC TGT TAT GGA AGG TTC ATG CCC CGG ATG GTT GTT 1304
 Arg Gly Val Ser Cys Tyr Gly Arg Phe Met Pro Arg Met Val Val
 415 420 425

ATT CCT CCT GGA ATG GAA TTC AAT CAT ATT GTT CCA CAT GAG GGT 1349
 Ile Pro Pro Gly Met Glu Phe Asn His Ile Val Pro His Glu Gly
 430 435 440

GAT ATG GAT GGT GAA ACA GAA GAA ACT GAA GAG CAT CCT ACA TCA 1394
 Asp Met Asp Gly Glu Thr Glu Glu Thr Glu Glu His Pro Thr Ser
 445 450 455

CCT GAT CCA CCT ATC TGG GCT GAG ATT ATG CGC TTC TTT TCT AAA 1439
 Pro Asp Pro Pro Ile Trp Ala Glu Ile Met Arg Phe Phe Ser Lys
 460 465 470

CCA AGG AAG CCA ATG ATA CTT GCC CTT GCT AGG CCT GAC CCG AAG 1484
 Pro Arg Lys Pro Met Ile Leu Ala Leu Ala Arg Pro Asp Pro Lys
 475 480 485

20

AAG	AAT	ATC	ACG	ACT	TTG	GTC	AAA	GCA	TTT	GGA	GAA	TGC	CGT	CCA	1529
Lys	Asn	Ile	Thr	Thr	Leu	Val	Lys	Ala	Phe	Gly	Glu	Cys	Arg	Pro	
					490				495					500	

CTA	AGG	GAG	CTA	GCT	AAT	CTT	ACT	CTT	ATA	ATG	GGT	AAC	CGA	GAT	1574
Leu	Arg	Glu	Leu	Ala	Asn	Leu	Thr	Leu	Ile	Met	Gly	Asn	Arg	Asp	
				505				510						515	

GGT	ATT	GAC	GAG	ATG	TCA	AGC	ACC	AGT	TCT	TCA	GTT	CTC	CTG	TCA	1619
Gly	Ile	Asp	Glu	Met	Ser	Ser	Thr	Ser	Ser	Ser	Val	Leu	Leu	Ser	
				520				525						530	

GTG	CTT	AAG	CTA	ATT	GAT	CAA	TAC	GAC	CTT	TAT	GGT	CAA	GTA	GCA	1664
Val	Leu	Lys	Leu	Ile	Asp	Gln	Tyr	Asp	Leu	Tyr	Gly	Gln	Val	Ala	
				535				540						545	

TAC	CCC	AAA	CAT	CAC	AAG	CAA	GCT	GAT	GTT	CCT	GAG	ATT	TAT	CGT	1709
Tyr	Pro	Lys	His	His	Lys	Gln	Ala	Asp	Val	Pro	Glu	Ile	Tyr	Arg	
				550				555						600	

TTG	GCA	GCA	AAG	ACA	AAG	GGA	GTC	TTT	ATT	AAT	CCA	GCT	TTT	ATT	1754
Leu	Ala	Ala	Lys	Thr	Lys	Gly	Val	Phe	Ile	Asn	Pro	Ala	Phe	Ile	
				605				610						615	

GAG	CCA	TTT	GGG	CTG	ACT	CTA	ATA	GAG	GCA	GCA	GCT	CAT	GGT	TTA	1799
Glu	Pro	Phe	Gly	Leu	Thr	Leu	Ile	Glu	Ala	Ala	Ala	His	Gly	Leu	
				620				625						630	

21

CCG ATG GTT GCT ACG AAA AAT GGA GGC CCT GTT GAT ATC CAG AGG 1844
 Pro Met Val Ala Thr Lys Asn Gly Gly Pro Val Asp Ile Gln Arg
 635 640 645

GTC CTT GAT AAT GGT CTT CTT GTG GAT CCT CAT GAG CAG CAG TCT 1889
 Val Leu Asp Asn Gly Leu Leu Val Asp Pro His Glu Gln Gln Ser
 650 655 660

ATT GCT ACT GCT TTG CTG AAG CTT GTT GCT GAT AAG CAA CTA TGG 1934
 Ile Ala Thr Ala Leu Leu Lys Leu Val Ala Asp Lys Gln Leu Trp
 665 670 675

ACA AAA TGC CAG CAA AAT GGA CTG AAA AAT ATT CAT CTC TAC TCT 1979
 Thr Lys Cys Gln Gln Asn Gly Leu Lys Asn Ile His Leu Tyr Ser
 680 685 690

TGG CCA GAG CAT TCG AAG ACA TAC CTA TCT CGA ATA GCC AGT TCG 2024
 Trp Pro Glu His Ser Lys Thr Tyr Leu Ser Arg Ile Ala Ser Ser
 695 700 705

AGA CAA AGG CAA CCA CAG TGG CAA AGA AGT AGT GAT GAA GGG CTT 2069
 Arg Gln Arg Gln Pro Gln Trp Gln Arg Ser Ser Asp Glu Gly Leu
 710 715 720

GAC AAT CAA GAG CCT GAA TCT CCA AGT GAT TCT TTA AGA GAT ATA 2114
 Asp Asn Gln Glu Pro Glu Ser Pro Ser Asp Ser Leu Arg Asp Ile
 725 730 735

22

AAG GAT ATA TCT CTA AAC CTT GAA GTT CTC GTT AGA CCG GAG AAA 2159
Lys Asp Ile Ser Leu Asn Leu Glu Val Leu Val Arg Pro Glu Lys
740 745 750

AGG GTG AAG ACG TTG AAA ATC TTG GGA TTG ATG ACA AAA GCA AAT 2204
Arg Val Lys Thr Leu Lys Ile Leu Gly Leu Met Thr Lys Ala Asn
755 760 765

TCG AGA ATG CTG TTA TGT TCA TGG TCT AAT GGT GTC CAT AAG ATG 2249
Ser Arg Met Leu Leu Cys Ser Trp Ser Asn Gly Val His Lys Met
770 775 780

CTT CGG AAG GCT CGG TTC TCT GAC AAA GTA GAT CAG GCT TCT AGT 2294
Leu Arg Lys Ala Arg Phe Ser Asp Lys Val Asp Gln Ala Ser Ser
785 790 795

AAA TAT CCA GCA TTT AGG AGG AGA AAA CTT ATA TAT GTT ATT GCT 2339
Lys Tyr Pro Ala Phe Arg Arg Arg Lys Leu Ile Tyr Val Ile Ala
800 805 810

GTA GAC GGG GAT TAT GAA GAT GGA CTT TTT GAT ATT GTT CGG AGG 2384
Val Asp Gly Asp Tyr Glu Asp Gly Leu Phe Asp Ile Val Arg Arg
815 820 825

ATA TTT GAT GCT GCT GGC AAG GAG AAG ATT GAA GGT TCC ATC GGG 2429
Ile Phe Asp Ala Ala Gly Lys Glu Lys Ile Glu Gly Ser Ile Gly
830 835 840

23

TTT ATA TTG TCA ACA TCC TAT TCT ATG CCC GAA ATT CAG AAC TAT 2474
 Phe Ile Leu Ser Thr Ser Tyr Ser Met Pro Glu Ile Gln Asn Tyr
 845 850 855

TTG CTA TCA AAA GGC TTC AAT CTT CAT GAT TTT GAT GCA TAT ATA 2519
 Leu Leu Ser Lys Gly Phe Asn Leu His Asp Phe Asp Ala Tyr Ile
 860 865 870

TGC AAC AGT GGG AGT GAG TTG TAC TAT TCA TCT TTG AAC TCA GAG 2564
 Cys Asn Ser Gly Ser Glu Leu Tyr Tyr Ser Ser Leu Asn Ser Glu
 875 880 885

GAG AGT AAT ATT ATA GCA GAT TCA GAT TAC CAT TCA CAC ATA GAG 2609
 Glu Ser Asn Ile Ile Ala Asp Ser Asp Tyr His Ser His Ile Glu
 890 895 900

TAC AGA TGG GGT GGA GAA GGC CTT AGA AGG ACT TTG CTT CGC TGG 2654
 Tyr Arg Trp Gly Gly Glu Gly Leu Arg Arg Thr Leu Leu Arg Trp
 905 910 915

GCA GCT TCC ATC ACA GAA AAA AAT GGT GAA AAC GAA GAA CAG GTT 2699
 Ala Ala Ser Ile Thr Glu Lys Asn Gly Glu Asn Glu Glu Gln Val
 920 925 930

ATT ACT GAA GAT GAA GAA GTT TCT ACG GGT TAT TGC TTT GCG TTT 2744
 Ile Thr Glu Asp Glu Glu Val Ser Thr Gly Tyr Cys Phe Ala Phe
 935 940 945

24

AAA	ATA	AAG	AAC	CAA	AAT	AAG	GTT	CCC	CCT	ACG	AAG	GAG	CTC	CGC	2789
Lys	Ile	Lys	Asn	Gln	Asn	Lys	Val	Pro	Pro	Thr	Lys	Glu	Leu	Arg	
				950				955						960	

AAG	TCA	ATG	AGG	ATT	CAA	GCT	CTT	CGT	TGC	CAT	GTG	ATT	TAC	TGT	2834
Lys	Ser	Met	Arg	Ile	Gln	Ala	Leu	Arg	Cys	His	Val	Ile	Tyr	Cys	
				965				970						975	

CAG	AAC	GGA	TCT	AAA	ATG	AAT	GTG	ATT	CCA	GTA	CTA	GCA	TCC	CGT	2879
Gln	Asn	Gly	Ser	Lys	Met	Asn	Val	Ile	Pro	Val	Leu	Ala	Ser	Arg	
				980				985						990	

TCT	CAA	GCC	CTC	AGG	TAT	CTT	TAT	GTT	CGT	TGG	GGA	GTT	GAG	TTG	2924
Ser	Gln	Ala	Leu	Arg	Tyr	Leu	Tyr	Val	Arg	Trp	Gly	Val	Glu	Leu	
				995				1000						1005	

TCG	AAG	ATG	GTT	GTC	TTT	GTT	GGA	GAA	TGT	GGT	GAC	ACA	GAT	TAT	2969
Ser	Lys	Met	Val	Val	Phe	Val	Gly	Glu	Cys	Gly	Asp	Thr	Asp	Tyr	
				1010				1015						1020	

GAA	GGC	TTG	CTT	GGC	GGG	GTC	CAT	AAA	ACC	GTA	ATA	CTG	AAG	GGA	3014
Glu	Gly	Leu	Leu	Gly	Gly	Val	His	Lys	Thr	Val	Ile	Leu	Lys	Gly	
				1025				1030						1035	

GTC	TCC	AAC	ACT	GCT	TTA	AGG	TCT	CTC	CAT	GCC	AAC	AGA	AGT	TAC	3059
Val	Ser	Asn	Thr	Ala	Leu	Arg	Ser	Leu	His	Ala	Asn	Arg	Ser	Tyr	
				1040				1045						1050	

25

CCT CTT TCA CAT GTC GTG TCG CTT GAC AGC CCC AAT ATT GGC GAG 3104
Pro Leu Ser His Val Val Ser Leu Asp Ser Pro Asn Ile Gly Glu
1055 1060 1065

GTG AGC AAA GGG TGC AGC AGC TCC GAG ATC CAG TCC ATC GTC ACA 3149
Val Ser Lys Gly Cys Ser Ser Ser Glu Ile Gln Ser Ile Val Thr
1070 1075 1080

AAA CTC TCC AAA GCT TAA TCAGATATCT GCTGCTTTCT TTTGGGTAAG 3197
Lys Leu Ser Lys Ala End
1085

CAAGGTTTCA TCTTATATGA TTATATCATA AGATACTATA TAAGCACCTT 3247

ATTGGTAAGT CAGTCCCATA ATAATAATGT ACTTCAGAAC CACAATACTT 3297

AAAAGTTGGT TCAGTAGTGA TTAGTCTCAT AATAATCATA TAATTACACA 3347

TCCGCTGTTA ACTAGTGGTA ATATCTAAGC TCAACAATAA AGATGTAAAA 3397

TGCTAGTATG GAAATGAATT GCTAGCTGTT GATCTCTTTC CCTTTATTCT 3447

GTATTATTTTCT TTTCTCATC TCATGTAAAA ACAATTTTCT GAAGGTGTAC 3497

AGTTTTTTTCC CCTTATATAT CTGTATTATT TCTACTATTT TTTGTTTGTA 3547

AGAATATCCT CTCATCGAGG AGTGATAATT AAATAACCGG CTTGCTAAAT 3597

ATAAGCTTA TTCGAGTTAA AAAAAAAAAA AAAAAAAAAA 3635

26

The sucrose-synthase has the following nucleotide sequence
(Seq. ID No. 4):

CT GCA GGA GGG AAA CAA ATT CTT AGC GAT GGC CCG TTT AGC GAA	0044
Ala Gly Gly Lys Gln Ile Leu Ser Asp Gly Pro Phe Ser Glu	
5 10	
GTT CTT AGG TCT GCT CAG GAA GCA ATA GTT GTT CCT CCC TTT GTT	0089
Val Leu Arg Ser Ala Gln Glu Ala Ile Val Val Pro Pro Phe Val	
15 20 25	
GCT ATA GCA GTC CGT CCA AGA CCT GGA GTT TGG GAA TAT GTT CGT	0134
Ala Ile Ala Val Arg Pro Arg Pro Gly Val Trp Glu Tyr Val Arg	
30 35 40	
GTT AAT GTC TCT GAA CTG AAT GTG GAG CAG CTA ACT GTG TCT GAG	0179
Val Asn Val Ser Glu Leu Asn Val Glu Gln Leu Thr Val Ser Glu	
45 50 55	
TAT CTC CAT TTC AAG GAA GAA CTT GTG GAT GGA AAG GCT GAT GAC	0224
Tyr Leu His Phe Lys Glu Glu Leu Val Asp Gly Lys Ala Asp Asp	
60 65 70	
CAC TAT GTG CTT GAG CTT GAT TTC GAG CCT TTT AAT GAA TCA GTT	0269
His Tyr Val Leu Glu Leu Asp Phe Glu Pro Phe Asn Glu Ser Val	
75 80 85	
CCA CGT CCA ACT CGC TCT TCA TCA ATT GGT AAT GGT GTT CAG TTC	0314
Pro Arg Pro Thr Arg Ser Ser Ser Ile Gly Asn Gly Val Gln Phe	
90 95 100	
CTC AAT CGG CAC CTG TCA TCA AGC ATG TTC TGC AAC AAA GAT TGC	0359
Leu Asn Arg His Leu Ser Ser Ser Met Phe Cys Asn Lys Asp Cys	
105 110 115	

27

TTG GAG CCG TTA CTT GAT TTT CTT AGA GTG CAC AAA CAT AAA GGA 0404
 Leu Glu Pro Leu Leu Asp Phe Leu Arg Val His Lys His Lys Gly
 120 125 130

GTT GTC ATG ATG TTG AAT GAT CGG ATA CAG ACT ATC CAG CGT CTT 0449
 Val Val Met Met Leu Asn Asp Arg Ile Gln Thr Ile Gln Arg Leu
 135 140 145

CAG TCT GCA TTG TCT AAA GCT GAG GAT TAT CTT ATC AAA CTT CCA 0494
 Gln Ser Ala Leu Ser Lys Ala Glu Asp Tyr Leu Ile Lys Leu Pro
 150 155 160

GCA GAT ACA CCT TAC TCT GAG TTC GAA TTT GTA ATC CAA GGT ATG 0539
 Ala Asp Thr Pro Tyr Ser Glu Phe Glu Phe Val Ile Gln Gly Met
 165 170 175

GGT TTT GAA AGA GGC TGG GGT GAT ACT GCT GAA AGG GTT CTA GAA 0584
 Gly Phe Glu Arg Gly Trp Gly Asp Thr Ala Glu Arg Val Leu Glu
 180 185 190

ATG ATG CAT CTA CTA CTA GAT ATC CTT CAG GCT CCC GAT CCG TCT 0629
 Met Met His Leu Leu Leu Asp Ile Leu Gln Ala Pro Asp Pro Ser
 195 200 205

ACA TTA GAG ACA TTT CTG GGA AGA CTT CCC ATG GTG TTT AAT GTG 0674
 Thr Leu Glu Thr Phe Leu Gly Arg Leu Pro Met Val Phe Asn Val
 210 215 220

28

GTC ATT TTG TCT GTA CAT GGA TAT TTT GGA CAG GCA CAT GTG CTC 0719
Val Ile Leu Ser Val His Gly Tyr Phe Gly Gln Ala His Val Leu
225 230 235

GGC TTG CCT GAC ACT GGT GGG CAG ATA GTT TAT ATA CTT GAC CAA 0764
Gly Leu Pro Asp Thr Gly Gly Gln Ile Val Tyr Ile Leu Asp Gln
240 245 250

GTG CGG TCT CTG GAA CAT GAA ATG CTC CAA CGA ATA AAG AAG CAA 0809
Val Arg Ser Leu Glu His Glu Met Leu Gln Arg Ile Lys Lys Gln
255 260 265

GGA CTA GAT GTG ACT CCT AGA ATT CTT ATC GTG AGT CGG TTG ATT 0854
Gly Leu Asp Val Thr Pro Arg Ile Leu Ile Val Ser Arg Leu Ile
270 275 280

CCT GAC GCT AAA GGG ACC ACG TGC AAT CAA CGT ATG GAG AAA GTC 0899
Pro Asp Ala Lys Gly Thr Thr Cys Asn Gln Arg Met Glu Lys Val
285 290 295

AGT GGA ACA GAG CAT GCT AGT ATC CTG AGA GTT CCT TTC CGA TCA 0944
Ser Gly Thr Glu His Ala Ser Ile Leu Arg Val Pro Phe Arg Ser
300 305 310

GAG AAA GGA ATC CTC CGC AAA TGG ATA TCT AGA TTT GAT GTA TGG 0989
Glu Lys Gly Ile Leu Arg Lys Trp Ile Ser Arg Phe Asp Val Trp
315 320 325

29

CCT TAT TTA GAG ACC TTC ACT GAG GAT GCA GCT GGT GAA ATT ATT 1034
 Pro Tyr Leu Glu Thr Phe Thr Glu Asp Ala Ala Gly Glu Ile Ile
 330 335 340

GGC GAG TTG CAG GGT CGT CCA GAT CTG ATA ATT GGC AAC TAC AGC 1079
 Gly Glu Leu Gln Gly Arg Pro Asp Leu Ile Ile Gly Asn Tyr Ser
 345 350 355

GAT GGG AAT ATA GTT GCT TCT TTA TTG TCC CAC AAA ATG GGT GTC 1124
 Asp Gly Asn Ile Val Ala Ser Leu Leu Ser His Lys Met Gly Val
 360 365 370

ACC CAG TGC AAT ATA GCC CAT GCA TTG GAG AAA ACC AAG TAT CCA 1169
 Thr Gln Cys Asn Ile Ala His Ala Leu Glu Lys Thr Lys Tyr Pro
 375 380 385

GAT TCT GAT ATT TAC TGG AAA AGA TTT GAG GAC AAA TAT CAC TTC 1214
 Asp Ser Asp Ile Tyr Trp Lys Arg Phe Glu Asp Lys Tyr His Phe
 390 395 400

TCG TGT CAA TTT TCA GCT GAC TTG ATG GCA ATG AAT CAT GCT GAT 1259
 Ser Cys Gln Phe Ser Ala Asp Leu Met Ala Met Asn His Ala Asp
 405 410 415

TTC ATC ATT ACG AGT ACT TAC CAA GAG ATA GCT GGA ACG AAG AAT 1304
 Phe Ile Ile Thr Ser Thr Tyr Gln Glu Ile Ala Gly Thr Lys Asn
 420 425 430

30

ACT	GTT	GGT	CAA	TAT	GAA	AGC	CAT	AAG	GCC	TTT	ACT	TTT	CCG	GGG	1349
Thr	Val	Gly	Gln	Tyr	Glu	Ser	His	Lys	Ala	Phe	Thr	Phe	Pro	Gly	
435					440					445					

CTG	TAT	CGG	GTG	GTT	CAC	GGG	ATT	GAT	GTC	TTT	GAT	CCC	AAG	TTT	1394
Leu	Tyr	Arg	Val	Val	His	Gly	Ile	Asp	Val	Phe	Asp	Pro	Lys	Phe	
450					455					460					

AAT	ATT	GTC	TCG	CCA	GGG	GCA	GAC	ATG	GCC	ATC	TAC	TTC	CCA	TTT	1439
Asn	Ile	Val	Ser	Pro	Gly	Ala	Asp	Met	Ala	Ile	Tyr	Phe	Pro	Phe	
465					470					475					

TCA	GAG	AAG	GAT	GTC	ACC	TGT	CTC	ACT	TCA	CTT	CAT	AGA	CTT	ATA	1484
Ser	Glu	Lys	Asp	Val	Thr	Cys	Leu	Thr	Ser	Leu	His	Arg	Leu	Ile	
480					485					490					

GAG	CAG	CTC	CTA	TTC	AAA	CCT	GAG	CAG	AAC	GAA	GAA	CAC	ATT	GGT	1529
Glu	Gln	Leu	Leu	Phe	Lys	Pro	Glu	Gln	Asn	Glu	Glu	His	Ile	Gly	
495					500					505					

GTA	TTA	GAT	GAT	ACC	TCA	AAG	CCA	ATT	ATA	TTT	TCC	ATG	GCG	AGG	1574
Val	Leu	Asp	Asp	Thr	Ser	Lys	Pro	Ile	Ile	Phe	Ser	Met	Ala	Arg	
510					515					520					

CTA	GAC	CGT	GTG	AAG	AAT	ATA	ACA	GGG	CTG	GTA	GAG	TGC	TAT	GGC	1619
Leu	Asp	Arg	Val	Lys	Asn	Ile	Thr	Gly	Leu	Val	Glu	Cys	Tyr	Gly	
525					530					535					

AAG AAT GCG AAA CTC AGG GAA CTG GCA AAC CTG GTT GTA GTG GCT 1664
Lys Asn Ala Lys Leu Arg Glu Leu Ala Asn Leu Val Val Val Ala
540 545 550

GGG TAC AAT GAT GTA AAA AAG TCG AAT GAC AGG GAG GAA ATT GCC 1709
Gly Tyr Asn Asp Val Lys Lys Ser Asn Asp Arg Glu Glu Ile Ala
555 560 565

GAA ATC GAG AAG ATG CAC AGG CTT ATA CAG GAG TAT AAT TTA AGA 1754
Glu Ile Glu Lys Met His Arg Leu Ile Gln Glu Tyr Asn Leu Arg
570 575 580

GGA CAA TTT CGC TGG ATT GCT TCT CAA ACA AAT AGA GTA CGA AAT 1799
Gly Gln Phe Arg Trp Ile Ala Ser Gln Thr Asn Arg Val Arg Asn
585 590 595

GGT GAA CTC TAT CGC TAC ATT TGT GAC AAA GGA GGT ATT TTT GCG 1844
Gly Glu Leu Tyr Arg Tyr Ile Cys Asp Lys Gly Gly Ile Phe Ala
600 605 610

CAG CCT GCA TTT TAT GAA GCA TTT GGG CTT ACA GTT GTT GAA GCC 1889
Gln Pro Ala Phe Tyr Glu Ala Phe Gly Leu Thr Val Val Glu Ala
615 620 625

ATG ACC TGT GGT CTT CCC ACA TTT GCT ACC TGC CAC GGT GGT CCA 1934
Met Thr Cys Gly Leu Pro Thr Phe Ala Thr Cys His Gly Gly Pro
630 635 640

32

GCT GAG ATT ATA GAA GAC GGT GTT TCA GGA TTT CAT ATC GAT CCA 1979
 Ala Glu Ile Ile Glu Asp Gly Val Ser Gly Phe His Ile Asp Pro
 645 650 655

TAT CAT GCT GAT CAG GCA GAA AAA ATG ACT GAA TTC TTT GTC AAG 2024
 Tyr His Ala Asp Gln Ala Glu Lys Met Thr Glu Phe Phe Val Lys
 660 665 670

TGC AGA GAG GAT CCA AAC TAC TGG ACT AAA ATC TCT GCA GGA GGG 2069
 Cys Arg Glu Asp Pro Asn Tyr Trp Thr Lys Ile Ser Ala Gly Gly
 675 680 685

TTA CTA AGG ATC AAA GAA AGA TAT ACC TGG CAA AAG TAT TCT GAA 2114
 Leu Leu Arg Ile Lys Glu Arg Tyr Thr Trp Glu Lys Tyr Ser Glu
 690 695 700

AGG TTA ATG ACA TTG GCA GGG GTG TAT GGT TTC TGG AAA TAT GTC 2159
 Arg Leu Met Thr Leu Ala Gly Val Tyr Gly Phe Trp Lys Tyr Val
 705 710 715

TCT AAA CTA GAG AGA AGA GAG ACA CGA CGT TAT CTT GAG ATG TTC 2204
 Ser Lys Leu Glu Arg Arg Glu Thr Arg Arg Tyr Leu Glu Met Phe
 720 725 730

TAC ATT TTG AAG TTC CGT GAT CTG GCC AAC TCT GTT CCG CTG GCA 2249
 Tyr Ile Leu Lys Phe Arg Asp Leu Ala Asn Ser Val Pro Leu Ala
 735 740 745

33

ACA GAT GAA GAG CCT TCT ACT ACT GAT GCA GTT GCG ACA TTC CGT 2294
 Thr Asp Glu Glu Pro Ser Thr Thr Asp Ala Val Ala Thr Phe Arg
 750 755 760

GGA CCT TGA ACGCTGCTGC TTACTGAGGT TCCAAGTTGT GTATATATTA. 2343
 Gly Pro End

CTGTGAAAGG AATAAGTGTA GCTACACAAA AGGTTCTCAA CTATTAGTAT 2393

CTTCTCTGTG TAAATAACGA GAGTGAAAAA TGTAATATTG TTGATGTCTT 2443

GAAAACTGAG TTTGCTTTGT TTATTTTAA GTGTATGACA ATATGTATCA 2493

TATAACGGAT TCTTCAGTGA TCATATCAAA AACTACTGAC CATCGAAGTT 2543

AATGAAAATC GACAGCAACA 2563

- 5 These sequences can also be combined together in a suitable plasmid which leads to a combination of the individual characteristics, conditioned by the expression of the protein.
- 10 The promoter should ensure that the foreign gene is expressed in the plant. The promoter can be so chosen that the expression occurs only in specified tissues, at a determined time point in the plant's development or at a time point determined by outside influences. The promoter
- 15 can be homologous or heterologous to the plant. Suitable promoters are e.g. the promoter of the 35S RNA of the cauliflower mosaic virus, the patatin promoter B33 (Rocha-Sosa et al. (1989) EMBO J 8: 23-29) or a promoter

that ensures an expression only in photosynthetically active tissues. Other promoters can be used which ensure an expression only in specified organs, such as the root, tuber, seed, stem or specified cell types such as
5 mesophyllic, epidermal or transport cells.

The coding sequences described herein contain the information for the formation of an mRNA for the large subunit of the ADP-glucose-pyrophosphorylase and the
10 sucrose-phosphate-synthase (SPS) and a part of the information for formation of the small subunit of the ADP-glucose-pyrophosphorylase as well as the sucrose-synthase, that are suitable for the formation of anti-sense RNA to the corresponding genes. Whether a
15 translatable mRNA or an anti-sense nucleic acid is formed, depends on the orientation of the coding sequence in relation to the promoter. If the 3' end of the coding sequence is fused to the 3' end of the promoter, an anti-sense RNA results, and by fusion of the 5' end of the
20 coding to the 3' end of the promoter a translatable RNA results. This latter leads to an increase of the enzyme activity in the cell, whilst the first leads to a reduction of the enzyme activity in the cell.

25 The coding sequence for the large and small subunit of the ADP-glucose-pyrophosphorylase, the sucrose phosphate synthase and the sucrose synthase can be one of those described in this invention or can be one that is derived by modifications of the sequences described above.
30 Thereby especially modifications of the sequences can be considered which lead to by-passing of the plant's own regulation mechanisms. Modifications to the DNA sequences of the invention can be by known methods, such as e.g. base exchange or targeted or non-targeted mutagenesis. The
35 so-formed derivatives of the DNA sequences of the invention are also within the scope of the invention.

With plasmids, which contain one or more of the DNA sequences of the invention, sugar beet can be transformed with the object of raising and/or reducing the enzyme activity and/or the change of the sucrose concentration.

5

For the introduction of the DNA sequences of the invention in sugar beet, a large number of cloning vectors are available, which contain a replication signal for *E. coli* and a marker, which allows a selection of the transformed cells.

10

According to the introduction method of the desired gene in the plant, other DNA sequences may be suitable. Should the Ti- or Ri-plasmid be used, e.g. for the transformation of the plant cell, then at least the right boundary, often however both the right and left boundary of the Ti- and Ri-Plasmid T-DNA, is attached, as a flanking region, to the gene being introduced. The use of T-DNA for the transformation of plants cells has been intensively researched and is well described in EP 120 516; Hoekama, In: The Binary Plant Vector System, Offset-drukkerij Kanters B.V. Alblasterdam, (1985), Chapter V; Fraley, et al., Crit. Rev. Plant Sci., 4:1-46 and An et al. (1985) EMBO J. 4: 277-287. Once the introduced DNA is integrated in the genome, it is as a rule stable there and remains also in the offspring of the original transformed cells. It normally contains a selection marker, which induces resistance in the transformed plant cells against a biocide or antibiotic such as kanamycin, G 418, bleomycin, hygromycin or phosphinotricin etc. The individual marker employed should therefore allow the selection of transformed cells from cells, which lack the introduced DNA.

35 For the introduction of DNA into a plant, besides transformation using *Agrobacteria*, there are many other.

techniques available. These techniques include the fusion of protoplasts, microinjection of DNA and electroporation, as well as ballistic methods and virus infection. From the transformed plant material, whole plants can be regenerated in a suitable medium, which contains antibiotics or biocides for the selection. The resulting plants can then be tested for the presence of introduced DNA. No special demands are placed on the plasmids in injection and electroporation. Simple plasmids, such as e.g. pUC-derivatives can be used. Should however whole plants be regenerated from such transformed cells the presence of a selectable marker gene is necessary. The transformed cells grow within the plants in the usual manner (see also McCormick et al. (1986) Plant Cell Reports 5: 81-84). These plants can be grown normally and crossed with plants, that possess the same transformed genes or different. The resulting hybrid individuals have the corresponding phenotypical properties.

20

In order to understand the examples forming the basis of this invention all the processes necessary for these tests and which are known per se will first of all be listed:

25

1. Cloning process

The vectors pUC 18/19 and M13mp10 series (Yanisch-Perron et al. (1985) Gene 33: 103-119), as well as the vector EMBL 3 (Frischauf et al. (1983) J Mol Biol 170: 827- 842) were used for cloning.

30

For the plant transformations, the gene constructs were cloned in the binary vector BIN 19 (Bevan (1984) Nucl. Acids Res 12: 8711-8720)

35

2. Bacterial strains

The *E. coli* strain BMH71-18 (Messing et al., Proc. Natl. Acad. Sci. USA (1977), 24, 6342-6346) or TB1 was used for the pUC and M13 mP vectors.

- 5 For the vector BIN19, the *E. coli* strain TB1 exclusively, was used. TB1 is a recombinant-negative, tetracycline-resistant derivative of strain JM101 (Yanisch-Perron et al., Gene (1985), 33, 103-119). The genotype of the TB1 strain is (Bart Barrel, personal communication):
- 10 F'(traD36, proAB, lacI, lacZΔM15), Δ(lac, pro), SupE, thiS, recA, Srl::Tn10(TcR).

The transformation of the plasmids into the potato plants was carried out using *Agrobacterium tumefaciens* strain

15 LBA4404 (Bevan, (1984), Nucl. Acids Res. 12, 8711-8720).

3. Transformation of *Agrobacterium tumefaciens*

- In the case of BIN19 derivatives, the insertion of the DNA
- 20 into the *Agrobacterium* was effected by direct transformation in accordance with the method of Holsters et al., (1978) (Mol Gene Genet 163: 181-187). The plasmid DNA of the transformed *Agrobacterium* was isolated in accordance with the method of Birnboim and Doly (1979)
- 25 (Nucl Acids Res 7: 1513-1523) and was analysed by gel electrophoresis after suitable restriction cleavage.

4. Sucrose phosphate-synthase activity test

- 30 The sucrose phosphate-synthase activity was determined according to the method of Siegel and Stitt (1990, Plant Science 66: 205-210) in a two stage analysis. To 180 μl of a solution of 50mM HEPES/KOH (pH 7.4), 5mM magnesium chloride, 5mM fructose-6-phosphate, 25mM
- 35 glucose-6-phosphate and 6mM uridine-5'-diphosphoglucose, 20 μl of probe was added and incubated for 10 minutes at

25°C. It was heated for 3 minutes at 95°C, to complete the reaction. After centrifuging, the supernatant was spectroscopically analysed for the liberation of uridine-5'-diphosphate, whereby a pyruvate-kinase coupling enzyme reaction was used. Preparations without hexose phosphate, as well as the measurement of the recovery of added uridine-5'-diphosphate act as controls.

Examples

Example 1

Cloning of cDNA to large and small subunits of the ADP glucose pyrophosphorylase of sugar beet.

From the storage roots of 3-4 month old sugar beet plants grown in the greenhouse, RNA was isolated according to the method of Logemann et al (1987, Anal Biochem 163, 16-20). Resulting from poly-A+-RNA, a cDNA library was laid down according to the method of Gubler and Hoffmann (1983, Gene 25, 263) in the expression vector Lambda Zap II XR. To this there was used an oligo-dT primer provided with an XhoI recognising position and for synthesis of the first cDNA strand methylated cytidine nucleotide was inserted. After synthesis of the two strands an EcoRI-adaptor was attached and removed to one side by again cutting with the restriction endonuclease XhoI. In this way the hemimethylation of cDNA was hindered so that an internal XhoI recognition position is cut. By these procedures there is obtained a population of cDNA molecules, that can be cloned directed into the EcoRI/XhoI cut DNA of the phage Lambda. After packing of recombinant phage-DNA in phage heads, 200000 plaque forming units of the bank were plated out for infection of a bacterial colony and then each is probed with the total cDNA fragment of the large and/or subunit of the AGPase of potato (Müller-Roeber et

al., 1990, MGG 224, 136-146) as an EcoRI-fragment. The recombinant phages corresponding to the hybridising signal were isolated. By *in vivo* excision plasmids were cut out from the Lambda zap-genome, which carry a double stranded
5 cDNA as insertion. The plasmids were transformed in bacterial cells. The plasmid-DNA was then propagated in the bacteria. After checking the size of the insertions individual clones were analysed by determination of the primary sequence.

10

Example 2

15 Cloning of cDNA to sucrose-phosphate-synthase (SPS) from sugar beet.

From the storage roots of 3-4 month old sugar beet plants grown in the greenhouse, RNA was isolated according to the method of Logemann et al (1987, Anal Biochem 163, 16-20).
20 Resulting from poly-A+-RNA, a cDNA library was laid down according to the method of Gubler and Hoffmann (1983, Gene 25, 263) in the expression vector Lambda Zap II XR. To this there was used an oligo-dT primer provided with an XhoI recognising position and for synthesis of the first
25 cDNA strand methylated cytidine nucleotide was inserted. After synthesis of the two strands an EcoRI-adaptor was attached and removed to one side by again cutting with the restriction endonuclease XhoI. In this way the hemimethylation of cDNA was hindered so that an internal
30 XhoI recognition position is cut. By these procedures there is obtained a population of cDNA molecules, that can be cloned directed into the EcoRI/XhoI cut DNA of the phage Lambda. After packing of recombinant phage-DNA in phage heads, 200000 plaque forming units of the bank were
35 plated out for infection of a bacterial colony and then each is probed with the total cDNA fragment of the

sucrose-phosphate-synthase (SPS) from spinach (Sonnewald, 1992, Planta) as NotI. The recombinant phages corresponding to the hybridising signal were isolated. By *in vivo* excision plasmids were cut out from the Lambda zap-genome, which carry a double stranded cDNA as insertion. The plasmids were transformed in bacterial cells. The plasmid-DNA was then propagated in the bacteria. After checking the size of the insertions individual clones were analysed by determination of the primary sequence.

Example 3

Cloning of cDNA to sucrose-synthase from sugar beet.

From the storage roots of 3-4 month old sugar beet plants grown in the greenhouse, RNA was isolated according to the method of Logemann et al (1987, Anal Biochem 163, 16-20). Resulting from poly-A⁺-RNA, a cDNA library was laid down according to the method of Gubler and Hoffmann (1983, Gene 25, 263) in the expression vector Lambda Zap II XR. To this there was used an oligo-dT primer provided with an XhoI recognising position and for synthesis of the first cDNA strand methylated cytidine nucleotide was inserted. After synthesis of the two strands an EcoRI-adaptor was attached and removed to one side by again cutting with the restriction endonuclease XhoI. In this way the hemimethylation of cDNA was hindered so that an internal XhoI recognition position is cut. By these procedures there is obtained a population of cDNA molecules, that can be cloned directed into the EcoRI/XhoI cut DNA of the phage Lambda. After packing of recombinant phage-DNA in phage heads, 200000 plaque forming units of the bank were plated out for infection of a bacterial colony and then parallel is probed with both EcoRI/BgIII sub fragments sucrose synthase from maize (Worrell et al., 1991, Plant

Cell 3, 1121-1130). The recombinant phages corresponding to the hybridising signal were isolated. By *in vivo* excision plasmids were cut out from the Lambda zap-genome, which carry a double stranded cDNA as insertion. The
5 plasmids were transformed in bacterial cells. The plasmid-DNA was then propagated in the bacteria. After checking the size of the insertions individual clones were analysed by determination of the primary sequence.

10 Example 4

Determination of the nucleotide sequence of the ADP
glucose pyrophosphorylase, the sucrose synthase and the
sucrose phosphate synthase of sugar beet and derivation of
15 the corresponding amino acid sequences

The nucleotide sequences of the insertions obtained from Examples 1 -3, were determined by standard methods by means of the dideoxy method (Sanger et al. (1977) Proc.
20 Natl. Acad. Sci. USA, 74, 5463-5467). The nucleotide sequences and the amino acid sequences derived therefrom are given in the sequence protocols Seq. ID No. 1-4.

The sequences are shown earlier; the protocols are as
25 follows:

42

SEQ ID NO: 1

SEQUENCE TYPE: Nucleotide with corresponding protein.

SEQUENCE LENGTH: 1924 base pairs

5 STRANDEDNESS: single

TOPOLOGY: linear

MOLECULE TYPE: cDNA

ORIGINAL SOURCE

10 ORGANISM: *Beta vulgaris*IMMEDIATE EXPERIMENTAL SOURCE: cDNA library in Phage Lamda
zap

FEATURES:

15 from 206 to 1770 coding region

PROPERTIES: ADP-glucose-pyrophosphorylase, large subunit

20 SEQ ID NO: 2

SEQUENCE TYPE: Nucleotide with corresponding protein

SEQUENCE LENGTH: 1763 base pairs

STRANDEDNESS: single

25 TOPOLOGY: linear

MOLECULE TYPE: cDNA

ORIGINAL SOURCE

ORGANISM: *Beta vulgaris*30 IMMEDIATE EXPERIMENTAL SOURCE: cDNA library in Phage Lamda
zap

FEATURES:

from 3 to 1469 coding region

35

PROPERTIES: ADP-glucose-pyrophosphorylase, small subunit

43

SEQ ID NO: 3

SEQUENCE TYPE: Nucleotide with corresponding protein

SEQUENCE LENGTH: 3635 base pairs

5 STRANDEDNESS: single

TOPOLOGY: linear

MOLECULE TYPE: cDNA

ORIGINAL SOURCE

10 ORGANISM: Beta vulgarisIMMEDIATE EXPERIMENTAL SOURCE: cDNA library in Phage Lamda
zap

FEATURES:

15 from 31 to 3164 coding region

PROPERTIES: Sucrose-phosphate-synthase

20 SEQ ID NO: 4

SEQUENCE TYPE: Nucleotide with corresponding protein

SEQUENCE LENGTH: 2563 base pairs

STRANDEDNESS: single

25 TOPOLOGY: linear

MOLECULE TYPE: cDNA

ORIGINAL SOURCE

ORGANISM: Beta vulgaris30 IMMEDIATE EXPERIMENTAL SOURCE: cDNA library in Phage Lamda
zap

FEATURES:

from 3 to 2300 coding region

35

PROPERTIES: Sucrose synthase

CLAIMS

1. DNA sequence with the coding region for the large subunit of ADP-glucose-pyrophosphorylase for the preparation of sugar beet with changed sucrose concentration, characterised in that the sequence has the following nucleotide sequence (Seq. ID No.1):

CAAAAGAAAA	ACTTCCCATT	TCTACTTCTT	TGCACAATAT	AATTTCCCAC	0050										
CAATTTTCT	TTAAATTTCT	CACTTTCATT	TAATCAGTTT	TCAGCAACAT	0100										
TCTGATACTC	GACAACCCAC	TTTCTGTTCT	CCCAAGATTC	CAAACCTCTG	0150										
ATTCTCATTC	CACTAATATT	TTTGCTTATT	TTTTTTCTGG	ATTTAAAGAA	0200										
AAGCT	ATG	GAT	GCA	AGT	GCA	GCA	GCC	ATA	AAT	GTC	AAT	GCC	CAT	0243	
	Met	Asp	Ala	Ser	Ala	Ala	Ala	Ile	Asn	Val	Asn	Ala	His		
					5					10					
TTA	ACA	GAA	GTT	GGA	AAG	AAA	CGT	TTT	TTA	GGA	GAG	AGA	ATC	AGT	0288
Leu	Thr	Glu	Val	Gly	Lys	Lys	Arg	Phe	Leu	Gly	Glu	Arg	Ile	Ser	
	15				20					25					
CAA	AGT	TTG	AAG	GGT	AAA	GAT	CTG	AGA	GCT	CTG	TTT	TCA	AGA	ACT	0333
Gln	Ser	Leu	Lys	Gly	Lys	Asp	Leu	Arg	Ala	Leu	Phe	Ser	Arg	Thr	
	30				35					40					
GAG	AGC	AAG	GGT	AGA	AAT	GTC	AAT	AAA	CCT	GGG	GTT	GCA	TTT	TCT	0378
Glu	Ser	Lys	Gly	Arg	Asn	Val	Asn	Lys	Pro	Gly	Val	Ala	Phe	Ser	
	45				50					55					
GTT	CTC	ACC	TCA	GAT	TTT	AAT	CAA	AGT	GTT	AAA	GAA	TCT	TTG	AAA	0423
Val	Leu	Thr	Ser	Asp	Phe	Asn	Gln	Ser	Val	Lys	Glu	Ser	Leu	Lys	
	60				65					70					

45

TAT GAG CCA GCA TTA TTT GAA TCT CCA AAA GCT GAC CCA AAA AAT 0468
 Tyr Glu Pro Ala Leu Phe Glu Ser Pro Lys Ala Asp Pro Lys Asn

75

80

85

GTG GCT GCA ATT GTG CTG GGT GGT GGT GCT GGG ACT CGC CTC TTT 0513
 Val Ala Ala Ile Val Leu Gly Gly Gly Ala Gly Thr Arg Leu Phe

90

95

100

CCT CTT ACT AGC AGG AGA GCT AAG CCA GCA GTG CCA ATT GGA GGG 0558
 Pro Leu Thr Ser Arg Arg Ala Lys Pro Ala Val Pro Ile Gly Gly

105

110

115

TGT TAC AGG CTG ATT GAT GTG CCT ATG AGC AAC TGC ATC AAC AGT 0603
 Cys Tyr Arg Leu Ile Asp Val Pro Met Ser Asn Cys Ile Asn Ser

120

125

130

GGC ATT AGA AAG ATT TTC ATT CTT ACC CAG TTC AAT TCG TTT TCG 0648
 Gly Ile Arg Lys Ile Phe Ile Leu Thr Gln Phe Asn Ser Phe Ser

135

140

145

CTT AAT CGT CAT CTT GCT CGA ACC TAT AAT TTT GGA GAT GGT GTG 0693
 Leu Asn Arg His Leu Ala Arg Thr Tyr Asn Phe Gly Asp Gly Val

150

155

160

AAT TTT GGG GAT GGC TTT GTG GAG GTT TTT GCT GCT ACA CAA ACA 0738
 Asn Phe Gly Asp Gly Phe Val Glu Val Phe Ala Ala Thr Gln Thr

165

170

175

46

CCT GGA GAA TCA GGA AAG AAA TGG TTC CAG GGC ACC GCT GAT GCA 0783
Pro Gly Glu Ser Gly Lys Lys Trp Phe Gln Gly Thr Ala Asp Ala
180 185 190

GTA AGA CAG TTT TTC TGG GCA TTT GAG GAT TCC AAA TCC AAG GAT 0828
Val Arg Gln Phe Phe Trp Ala Phe Glu Asp Ser Lys Ser Lys Asp
195 200 205

GTC GAG CAT ATA GTT ATT TTA TCC GGT GAT CAT CTT TAC CGA ATG 0873
Val Glu His Ile Val Ile Leu Ser Gly Asp His Leu Tyr Arg Met
210 215 220

GAT TAC ATG AGT TTT TGG CAG AAG CAC ATT GAC ACC AAT GCT GAT 0918
Asp Tyr Met Ser Phe Trp Gln Lys His Ile Asp Thr Asn Ala Asp
225 230 235

ATT ACA GTG TCA TGC ATA CCC ATG GAT GAC AGC CGT GCA TCG GAT 0963
Ile Thr Val Ser Cys Ile Pro Met Asp Asp Ser Arg Ala Ser Asp
240 245 250

TAT GGG CTG ATG AAG ATT GAT CAC ACT GGA CGC ATT GTC CAT TTT 1008
Tyr Gly Leu Met Lys Ile Asp His Thr Gly Arg Ile Val His Phe
255 260 265

GCA GAA AAA CCC AAG GGT TCT GAT CTA ACA GCA ATG CAA GTA GAT 1053
Ala Glu Lys Pro Lys Gly Ser Asp Leu Thr Ala Met Gln Val Asp
270 275 280

47

ACA ACT GTT CTT GGG CTC TCT GAC CTT GAA GCT ATG TCA AAT CCA 1098
 Thr Thr Val Leu Gly Leu Ser Asp Leu Glu Ala Met Ser Asn Pro
 285 290 295

TAT ATT GCA TCA ATG GGT GTT TAT GTC TTT CGA ACG GAT GTT CTT 1143
 Tyr Ile Ala Ser Met Gly Val Tyr Val Phe Arg Thr Asp Val Leu
 300 305 310

ATG GAG CTT CTC AAT CGA AAA TAC CCT TCA AGC AAT GAT TTT GGC 1188
 Met Glu Leu Leu Asn Arg Lys Tyr Pro Ser Ser Asn Asp Phe Gly
 315 320 325

TCT GAG ATT ATT CCT TCA GCT GTA GGA GAG TCT AAT GTT CAG GCA 1233
 Ser Glu Ile Ile Pro Ser Ala Val Gly Glu Ser Asn Val Gln Ala
 330 335 340

TAT CTA TTT AAT GAC TAC TGG GAG GAT ATC GGA ACC ATA AAG TCT 1278
 Tyr Leu Phe Asn Asp Tyr Trp Glu Asp Ile Gly Thr Ile Lys Ser
 345 350 355

TTC TTT GAT TCC AAT TTG GCC CTT ACA CAA CAG CCT CCC AAG TTT 1323
 Phe Phe Asp Ser Asn Leu Ala Leu Thr Gln Gln Pro Pro Lys Phe
 360 365 370

GAA TTC TAC GAT CCA AAA ACA CCT TTT TAT ACA TCT GCA AGA TTT 1368
 Glu Phe Tyr Asp Pro Lys Thr Pro Phe Tyr Thr Ser Ala Arg Phe
 375 380 385

48

CTG CCT CCT ACA AAA GTC GAC AGG TGC AAG ATT GTC GAT TCC ATT 1413
 Leu Pro Pro Thr Lys Val Asp Arg Cys Lys Ile Val Asp Ser Ile
 390 395 400

GTA TCC CAT GGT TGT TTT CTA CAG GAG TCT AGC ATC CAA CAT TCC 1458
 Val Ser His Gly Cys Phe Leu Gln Glu Ser Ser Ile Gln His Ser
 405 410 415

ATT GTT GGT GTT CGC TCA AGA TTA GAG TCC GGG GTT GAG TTC CAG 1503
 Ile Val Gly Val Arg Ser Arg Leu Glu Ser Gly Val Glu Phe Gln
 420 425 430

GAC ACC ATG ATG ATG GGC GCA GAT TAC TAT CAA ACT GAA TCA GAA 1548
 Asp Thr Met Met Met Gly Ala Asp Tyr Tyr Gln Thr Glu Ser Glu
 435 440 445

ATT GCT TCT CTG CTT GCT GAG GGA AAG GTT CCT GTT GGT GTC GGA 1593
 Ile Ala Ser Leu Leu Ala Glu Gly Lys Val Pro Val Gly Val Gly
 450 455 460

CAG AAT ACC AAA ATA AAG AAT TGC ATA ATT GAC AAG AAC GCC AAA 1638
 Gln Asn Thr Lys Ile Lys Asn Cys Ile Ile Asp Lys Asn Ala Lys
 465 470 475

ATT GGA AAA GAT GTG GTA ATC GCA AAC ACG GAT GGT GTT GAG GAA 1683
 Ile Gly Lys Asp Val Val Ile Ala Asn Thr Asp Gly Val Glu Glu
 480 485 490

49

GCA GAT AGA CCA AAT GAA GGC TTT TAC ATC AGG TCG GGC ATT ACC 1728
 Ala Asp Arg Pro Asn Glu Gly Phe Tyr Ile Arg Ser Gly Ile Thr
 495 500 505

ATC ATT TTG AAG AAC GCA ACC ATA CAA GAC GGT CTT GTG ATT TAG 1773
 Ile Ile Leu Lys Asn Ala Thr Ile Gln Asp Gly Leu Val Ile End
 510 515 520

ATTTAATCAT AACCTCATTA GAAAGAAATA ATTTTGCATG ATTTCCTTTT 1823

CATGTAACCT AAACCTGGCTA AACCACGAGG TTTTCTCATC TGTATATATA 1873

ATATGTCTAT AACTATGGAT AATCTTAATA AAAAAAAAAA AAAAAAAAAA 1923

A 1924

2. DNA sequence with the coding region for the small subunit of the ADP-glucose-pyrophosphatase for the preparation of sugar beet with changed sucrose concentration, characterised in that the sequence has the following nucleotide sequence (Seq. ID No.2):

GG ATA ACT GTG CCA TCA ACC TCC TCA AAG AAC CTC CAA AAT AGC 0044
 Ile Thr Val Pro Ser Thr Ser Ser Lys Asn Leu Gln Asn Ser
 5 10

CTC GCA TTC TCC TCT TCT TCT CTC TCC GGC GAC AAA ATT CAA ACG 0089
 Leu Ala Phe Ser Ser Ser Ser Leu Ser Gly Asp Lys Ile Gln Thr
 15 20 25

ACG TCA TTT CTC AAC CGC CGA TAT TGT AGA ATC TCT TCT AGA GCT 0134
 Thr Ser Phe Leu Asn Arg Arg Tyr Cys Arg Ile Ser Ser Arg Ala
 30 35 40

50

CCG	ATT	GTT	GTC	TCT	CCC	AAA	GCT	GTT	TCT	GAT	TCT	AAG	AAT	TCG	0179
Pro	Ile	Val	Val	Ser	Pro	Lys	Ala	Val	Ser	Asp	Ser	Lys	Asn	Ser	
45					50					55					

CAG	ACT	TGT	CTT	GAC	CCT	GAA	GCC	AGC	CGT	AGT	GTT	CTT	GGT	ATT	0224
Gln	Thr	Cys	Leu	Asp	Pro	Glu	Ala	Ser	Arg	Ser	Val	Leu	Gly	Ile	
60					65					70					

ATA	CTT	GGA	GGT	GGT	GCT	GGT	ACA	CGT	CTT	TAC	CCG	TTG	ACT	AAG	0269
Ile	Leu	Gly	Gly	Gly	Ala	Gly	Thr	Arg	Leu	Tyr	Pro	Leu	Thr	Lys	
75					80					85					

AAG	AGA	GCC	AAA	CCA	GCC	GTG	CCA	CTC	GGT	GCT	AAT	TAT	AGG	CTT	0314
Lys	Arg	Ala	Lys	Pro	Ala	Val	Pro	Leu	Gly	Ala	Asn	Tyr	Arg	Leu	
90					95					100					

ATT	GAT	ATC	CCA	GTG	AGC	AAT	TGT	TTG	AAC	AGT	AAT	ATT	TCC	AAA	0359
Ile	Asp	Ile	Pro	Val	Ser	Asn	Cys	Leu	Asn	Ser	Asn	Ile	Ser	Lys	
105					110					110					

ATA	TAT	GTT	CTT	ACA	CAA	TTC	AAT	TCT	GCT	TCT	CTG	AAT	CGT	CAT	0404
Ile	Tyr	Val	Leu	Thr	Gln	Phe	Asn	Ser	Ala	Ser	Leu	Asn	Arg	His	
115					120					125					

CTT	TCG	CGG	GCA	TAT	GCT	AGC	AAC	ATG	GGA	GGA	TAC	AAA	AAT	GAG	0449
Leu	Ser	Arg	Ala	Tyr	Ala	Ser	Asn	Met	Gly	Gly	Tyr	Lys	Asn	Glu	
130					135					140					

GGG	TTT	GTA	GAA	GTT	CTT	GCT	GCT	CAG	CAA	AGT	CCA	GAG	AAT	CCA	0494
Gly	Phe	Val	Glu	Val	Leu	Ala	Ala	Gln	Gln	Ser	Pro	Glu	Asn	Pro	
145					150					155					

51

AAC TGG TTT CAG GGT ACA GCT GAT GCT GTT AGG CAA TAT CTG TGG 0539
 Asn Trp Phe Gln Gly Thr Ala Asp Ala Val Arg Gln Tyr Leu Trp
 160 165 170

CTT TTC GAA GAG CAC AAT GTT CTT GAG TAC TTG ATT CTT GCT GGT 0584
 Leu Phe Glu Glu His Asn Val Leu Glu Tyr Leu Ile Leu Ala Gly
 175 180 185

GAC CAT TTG TAT CGA ATG GAT TAT GAA AGA TTT GTC CAA GCT CAC 0629
 Asp His Leu Tyr Arg Met Asp Tyr Glu Arg Phe Val Gln Ala His
 190 195 200

AGA GAA ACT GAT GCA GAC ATT ACT GTT GCT GCA TTG CCA ATG GAT 0674
 Arg Glu Thr Asp Ala Asp Ile Thr Val Ala Ala Leu Pro Met Asp
 205 210 215

GAA AAG CGT GCT ACT GCA TTT GGT TTG ATG AAA ATT GAT GAA GAA 0719
 Glu Lys Arg Ala Thr Ala Phe Gly Leu Met Lys Ile Asp Glu Glu
 220 225 230

GGA AGA ATT ATT GAG TTT GCC GAG AAA CCG AAA GGA GAA CAA TTG 0764
 Gly Arg Ile Ile Glu Phe Ala Glu Lys Pro Lys Gly Glu Gln Leu
 235 240 245

AAA GCT ATG AAG GTT GAT ACC ACA ATC CTG GGT CTG GAC GAT GAG 0809
 Lys Ala Met Lys Val Asp Thr Thr Ile Leu Gly Leu Asp Asp Glu
 250 255 260

52

AGA	GCA	AAA	GAA	ATG	CCA	TTC	ATA	GCC	AGC	ATG	GGC	ATA	TAT	GTT	0854
Arg	Ala	Lys	Glu	Met	Pro	Phe	Ile	Ala	Ser	Met	Gly	Ile	Tyr	Val	
265					270					275					

ATT	AGC	AAA	GAT	GTA	ATG	CTT	AAT	CTG	CTT	CGG	GAG	CAA	TTT	CCT	0899
Ile	Ser	Lys	Asp	Val	Met	Leu	Asn	Leu	Leu	Arg	Glu	Gln	Phe	Pro	
280					285					290					

GGT	GCT	AAT	GAT	TTT	GGA	AGT	GAA	GTT	ATT	CCA	GGC	GCC	ACT	TCC	0944
Gly	Ala	Asn	Asp	Phe	Gly	Ser	Glu	Val	Ile	Pro	Gly	Ala	Thr	Ser	
295					300					305					

ATA	GGG	TTG	AGA	GTC	CAA	GCT	TAT	TTG	TAT	GAT	GGT	TAC	TGG	GAG	0989
Ile	Gly	Leu	Arg	Val	Gln	Ala	Tyr	Leu	Tyr	Asp	Gly	Tyr	Trp	Glu	
310					315					320					

GAT	ATT	GGT	ACC	ATT	GAA	GCT	TTT	TAC	AAT	GCT	AAC	TTG	GGA	ATC	1054
Asp	Ile	Gly	Thr	Ile	Glu	Ala	Phe	Tyr	Asn	Ala	Asn	Leu	Gly	Ile	
325					330					335					

ACC	AAA	AAG	CCG	GTG	CCA	GAT	TTT	AGC	TTC	TAT	GAT	CGT	TCA	TCT	1079
Thr	Lys	Lys	Pro	Val	Pro	Asp	Phe	Ser	Phe	Tyr	Asp	Arg	Ser	Ser	
340					345					350					

CCA	ATT	TAT	ACA	CAA	CCT	CGG	TAT	TTG	CCT	CCT	TCA	AAG	ATG	CTT	1124
Pro	Ile	Tyr	Thr	Gln	Pro	Arg	Tyr	Leu	Pro	Pro	Ser	Lys	Met	Leu	
355					360					365					

53

GAT GCT GAT ATA ACT GAC AGC GTC ATC GGT GAA GGC TGT GTT ATT 1169
 Asp Ala Asp Ile Thr Asp Ser Val Ile Gly Glu Gly Cys Val Ile
 370 375 380

AAG AAC TGT AAG ATT CAT CAT TCT GTT ATC GGA CTT CGA TCT TGT 1214
 Lys Asn Cys Lys Ile His His Ser Val Ile Gly Leu Arg Ser Cys
 385 390 395

ATC TCG GAG GGT GCA ATC ATT GAG GAC ACA CTG TTG ATG GGA GCT 1259
 Ile Ser Glu Gly Ala Ile Ile Glu Asp Thr Leu Leu Met Gly Ala
 400 405 410

GAT TAT TAT GAG ACT GAT GCT GAT CGG AAA TTC CTG GCT GCT AAG 1304
 Asp Tyr Tyr Glu Thr Asp Ala Asp Arg Lys Phe Leu Ala Ala Lys
 415 420 425

GGT AGT GTA CCT ATT GGA ATT GGG AAT GCA CGT ATT GGG GAT GAT 1349
 Gly Ser Val Pro Ile Gly Ile Gly Asn Ala Arg Ile Gly Asp Asp
 430 435 440

GTC AAG ATT ATC AAC AGT GAC AAT GTA CAA GAA GCA GCA AGA GAA 1394
 Val Lys Ile Ile Asn Ser Asp Asn Val Gln Glu Ala Ala Arg Glu
 445 450 455

ACA GAC GGA TAC TTC ATA AAG AGC GGA ATA GTC ACT ATA ATC AAG 1439
 Thr Asp Gly Tyr Phe Ile Lys Ser Gly Ile Val Thr Ile Ile Lys
 460 465 470

54

GAC GCC ATG ATT CCA AGT GGA ACT GTA ATC TAG AAATGGAGCA 1482
 Asp Ala Met Ile Pro Ser Gly Thr Val Ile End
 475 480 485

TATAATAAAT ATCACTGCCT ATTTACAGTA CCTATCTGAG TCTCCCACCA 1532

TGACCCTTTG ATTCAATCTT TTAGTTATGT AAATATTTTT GGCTTTTGCG 1582

ATTTTGCCAT AAATTTGAAG AAGCGAGGAT TCAGGGACGA TAGTGCTATG 1632

AATTGGAAGA AAGGATTTGG GGGATATCTT TGTAAGACA TTTTGACTAC 1682

TGGGCACTAA AAATTTGGTA ATGCTATACC AAAATATATA AAAAGATCTT 1732

GCTGGGTTTT GTAAAAAAA AAAAAAAAAA A 1763

3. DNA sequence with the coding region for sucrose phosphate - synthase for the preparation of sugar beet with changed sucrose concentration, characterised in that the sequence has the following nucleotide sequence (Seq. ID No.3):

GGGCTGCAGG GAAGCTCTGA ACTTCAAAA ATG GCG GGA AAT GAT 0044
 Met Ala Gly Asn Asp

5

TGG ATA AAC AGT TAT TTA GAG GCA ATT CTG GAT GTG GGT CCA GGA 0089
 Trp Ile Asn Ser Tyr Leu Glu Ala Ile Leu Asp Val Gly Pro Gly

10

15

20

CTT GAT GAT GCA AAA TCA TCT TTG CTT TTG AGA GAA AGA GGC AGG 0134
 Leu Asp Asp Ala Lys Ser Ser Leu Leu Leu Arg Glu Arg Gly Arg

25

30

35

55

TTT	AGT	CCT	ACT	CGT	TAC	TTT	GTT	GAA	GAA	GTT	ATC	ACT	GGT	TTT	0179
Phe	Ser	Pro	Thr	Arg	Tyr	Phe	Val	Glu	Glu	Val	Ile	Thr	Gly	Phe	
				40					45					50	
GAT	GAA	ACC	GAC	CTT	CAT	CGT	TCA	TGG	GTT	CGG	GCA	CAA	GCA	ACA	0224
Asp	Glu	Thr	Asp	Leu	His	Arg	Ser	Trp	Val	Arg	Ala	Gln	Ala	Thr	
				55					60					65	
AGG	AGT	CCT	CAA	GAG	AGG	AAT	ACT	AGA	TTG	GAG	AAC	ATG	TGT	TGG	0269
Arg	Ser	Pro	Gln	Glu	Arg	Asn	Thr	Arg	Leu	Glu	Asn	Met	Cys	Trp	
				70					75					80	
AGA	ATT	TGG	AAT	TTG	GCT	CGT	CAG	AAG	AAG	CAG	CTT	GAG	AAT	GAA	0314
Arg	Ile	Trp	Asn	Leu	Ala	Arg	Gln	Lys	Lys	Gln	Leu	Glu	Asn	Glu	
				85					90					95	
GAA	GCT	CAG	CGG	AAG	ACA	AAA	CGT	CGT	ATG	GAG	CTT	GAG	AGG	GGT	0359
Glu	Ala	Gln	Arg	Lys	Thr	Lys	Arg	Arg	Met	Glu	Leu	Glu	Arg	Gly	
				100					105					110	
CGT	CGA	GAA	GCA	ACT	GCT	GAT	ATG	TCG	GAG	GAC	TTA	TCA	GAA	GGC	0404
Arg	Arg	Glu	Ala	Thr	Ala	Asp	Met	Ser	Glu	Asp	Leu	Ser	Glu	Gly	
				115					120					125	
GAA	AAG	GAC	ATT	TCA	GCT	CAT	GGT	GAT	AGC	ACC	CGT	CCT	AGA	TTG	0449
Glu	Lys	Asp	Ile	Ser	Ala	His	Gly	Asp	Ser	Thr	Arg	Pro	Arg	Leu	
				130					135					140	
CCA	AGA	ATA	AAT	TCT	CTT	GAT	GCT	ATG	GAG	ACA	TGG	ATT	AGT	CAA	0494
Pro	Arg	Ile	Asn	Ser	Leu	Asp	Ala	Met	Glu	Thr	Trp	Ile	Ser	Gln	
				145					150					155	

56

CAA	AAG	GAA	AAA	AAA	CTC	TAC	CTT	GTT	TTG	ATA	AGT	CTT	CAT	GGT	0539
Gln	Lys	Glu	Lys	Lys	Leu	Tyr	Leu	Val	Leu	Ile	Ser	Leu	His	Gly	
				160					165					170	

TTG	ATA	CGA	GGT	GAA	AAC	ATG	GAA	CTT	GGC	CGT	GAT	TCT	GAT	ACT	0584
Leu	Ile	Arg	Gly	Glu	Asn	Met	Glu	Leu	Gly	Arg	Asp	Ser	Asp	Thr	
				175					180					185	

GGT	GGT	CAG	GTT	AAG	TAT	GTG	GTT	GAG	CTT	GCA	AGG	GCT	CTA	GGT	0629
Gly	Gly	Gln	Val	Lys	Tyr	Val	Val	Glu	Leu	Ala	Arg	Ala	Leu	Gly	
				190					195					200	

TCG	ATG	CCA	GGT	GTT	TAT	AGA	GTT	GAT	TTG	CTA	ACT	AGG	CAA	GTT	0674
Ser	Met	Pro	Gly	Val	Tyr	Arg	Val	Asp	Leu	Leu	Thr	Arg	Gln	Val	
				205					210					215	

TCA	TCT	CCT	GAC	GTG	GAT	TGG	AGT	TAT	GGG	GAG	CCT	ACT	GAG	ATG	0719
Ser	Ser	Pro	Asp	Val	Asp	Trp	Ser	Tyr	Gly	Glu	Pro	Thr	Glu	Met	
				220					225					230	

CTG	AAT	CCA	AGG	GAT	TCC	AAT	GGT	TTT	GAT	GAT	GAT	GAT	GAT	GAA	0764
Leu	Asn	Pro	Arg	Asp	Ser	Asn	Gly	Phe	Asp	Asp	Asp	Asp	Asp	Glu	
				235					240					245	

ATG	GGA	GAG	AGT	AGT	GGT	GCT	TAC	ATT	GTT	CGT	ATA	CCA	TTT	GGG	0809
Met	Gly	Glu	Ser	Ser	Gly	Ala	Tyr	Ile	Val	Arg	Ile	Pro	Phe	Gly	
					250				255					260	

CCG	AGG	GAT	AAG	TAT	ATC	GCA	AAA	GAA	GAG	CTT	TGG	CCC	TAT	ATT	0854
Pro	Arg	Asp	Lys	Tyr	Ile	Ala	Lys	Glu	Glu	Leu	Trp	Pro	Tyr	Ile	
				265					270					275	

CCT	GAA	TTT	GTT	GAT	GGT	GCT	CTA	AAC	CAC	ATA	GTT	CAA	ATG	TCC	0899
Pro	Glu	Phe	Val	Asp	Gly	Ala	Leu	Asn	His	Ile	Val	Gln	Met	Ser	
				280				285					290		

AAA GTT TTA GGT GAG CAA ATT GGT AGC GGG GAA ACA GTT TGG CCA 0944
Lys Val Leu Gly Glu Gln Ile Gly Ser Gly Glu Thr Val Trp Pro
295 300 305

GTT GCC ATT CAT GGA CAT TAT GCT GAT GCT GGT GAT TCT GCT GCT 0989
Val Ala Ile His Gly His Tyr Ala Asp Ala Gly Asp Ser Ala Ala
310 315 320

CTT CTT TCT GGT GGC CTA AAT GTT CCA ATG CTT TTA ACG GGG CAT 1034
Leu Leu Ser Gly Gly Leu Asn Val Pro Met Leu Leu Thr Gly His
325 330 335

TCT	CTT	GGC	CGA	GAC	AAG	TTA	GAG	CAG	CTC	CTC	AAA	CAG	GGT	CGA	1079
Ser	Leu	Gly	Arg	Asp	Lys	Leu	Glu	Gln	Leu	Leu	Lys	Gln	Gly	Arg	
				340				345					350		

ATG TCT AAA GAT GAC ATA AAC AAT ACA TAC AAA ATA ATG CGT AGG 1124
Met Ser Lys Asp Asp Ile Asn Asn Thr Tyr Lys Ile Met Arg Arg
355 360 365

58

ATA	GAA	GCC	GAA	GAG	TTA	TCA	CTT	GAT	GCC	TCT	GAG	ATA	GTC	ATA	1169
Ile	Glu	Ala	Glu	Glu	Leu	Ser	Leu	Asp	Ala	Ser	Glu	Ile	Val	Ile	
				370					375					380	

ACT	AGT	ACA	AGA	CAA	GAA	ATA	GAA	GAG	CAA	TGG	CAC	CTC	TAT	GAT	1214
Thr	Ser	Thr	Arg	Gln	Glu	Ile	Glu	Glu	Gln	Trp	His	Leu	Tyr	Asp	
				385					390					395	

GGG	TTT	GAT	CCT	GTG	CTA	GAA	CGT	AAA	CTC	CGT	GCT	AGG	ATG	AAG	1259
Gly	Phe	Asp	Pro	Val	Leu	Glu	Arg	Lys	Leu	Arg	Ala	Arg	Met	Lys	
				400					405					410	

CGT	GGT	GTA	AGC	TGT	TAT	GGA	AGG	TTC	ATG	CCC	CGG	ATG	GTT	GTT	1304
Arg	Gly	Val	Ser	Cys	Tyr	Gly	Arg	Phe	Met	Pro	Arg	Met	Val	Val	
				415					420					425	

ATT	CCT	CCT	GGA	ATG	GAA	TTC	AAT	CAT	ATT	GTT	CCA	CAT	GAG	GGT	1349
Ile	Pro	Pro	Gly	Met	Glu	Phe	Asn	His	Ile	Val	Pro	His	Glu	Gly	
				430					435					440	

GAT	ATG	GAT	GGT	GAA	ACA	GAA	GAA	ACT	GAA	GAG	CAT	CCT	ACA	TCA	1394
Asp	Met	Asp	Gly	Glu	Thr	Glu	Glu	Thr	Glu	Glu	His	Pro	Thr	Ser	
				445					450					455	

CCT	GAT	CCA	CCT	ATC	TGG	GCT	GAG	ATT	ATG	CGC	TTC	TTT	TCT	AAA	1439
Pro	Asp	Pro	Pro	Ile	Trp	Ala	Glu	Ile	Met	Arg	Phe	Phe	Ser	Lys	
				460					465					470	

CCA	AGG	AAG	CCA	ATG	ATA	CTT	GCC	CTT	GCT	AGG	CCT	GAC	CCG	AAG	1484
Pro	Arg	Lys	Pro	Met	Ile	Leu	Ala	Leu	Ala	Arg	Pro	Asp	Pro	Lys	
				475					480					485	

CTA AGG GAG CTA GCT AAT CTT ACT CTT ATA ATG GGT AAC CGA GAT 1574
Leu Arg Glu Leu Ala Asn Leu Thr Leu Ile Met Gly Asn Arg Asp
505 510 515

GTG CTT AAG CTA ATT GAT CAA TAC GAC CTT TAT GGT CAA GTA GCA 1664
Val Leu Lys Leu Ile Asp Gln Tyr Asp Leu Tyr Gly Gln Val Ala
535 540 545

TTG GCA GCA AAG ACA AAG GGA GTC TTT ATT AAT CCA GCT TTT ATT 1754
Leu Ala Ala Lys Thr Lys Gly Val Phe Ile Asn Pro Ala Phe Ile
605 610 615

60

GAG CCA TTT GGG CTG ACT CTA ATA GAG GCA GCA GCT CAT GGT TTA 1799
 Glu Pro Phe Gly Leu Thr Leu Ile Glu Ala Ala Ala His Gly Leu
 620 625 630

CCG ATG GTT GCT ACG AAA AAT GGA GGC CCT GTT GAT ATC CAG AGG 1844
 Pro Met Val Ala Thr Lys Asn Gly Gly Pro Val Asp Ile Gln Arg
 635 640 645

GTC CTT GAT AAT GGT CTT CTT GTG GAT CCT CAT GAG CAG CAG TCT 1889
 Val Leu Asp Asn Gly Leu Leu Val Asp Pro His Glu Gln Gln Ser
 650 655 660

ATT GCT ACT GCT TTG CTG AAG CTT GTT GCT GAT AAG CAA CTA TGG 1934
 Ile Ala Thr Ala Leu Leu Lys Leu Val Ala Asp Lys Gln Leu Trp
 665 670 675

ACA AAA TGC CAG CAA AAT GGA CTG AAA AAT ATT CAT CTC TAC TCT 1979
 Thr Lys Cys Gln Gln Asn Gly Leu Lys Asn Ile His Leu Tyr Ser
 680 685 690

TGG CCA GAG CAT TCG AAG ACA TAC CTA TCT CGA ATA GCC AGT TCG 2024
 Trp Pro Glu His Ser Lys Thr Tyr Leu Ser Arg Ile Ala Ser Ser
 695 700 705

AGA CAA AGG CAA CCA CAG TGG CAA AGA AGT AGT GAT GAA GGG CTT 2069
 Arg Gln Arg Gln Pro Gln Trp Gln Arg Ser Ser Asp Glu Gly Leu
 710 715 720

61

GAC AAT CAA GAG CCT GAA TCT CCA AGT GAT TCT TTA AGA GAT ATA 2114
Asp Asn Gln Glu Pro Glu Ser Pro Ser Asp Ser Leu Arg Asp Ile
725 730 735

AAG GAT ATA TCT CTA AAC CTT GAA GTT CTC GTT AGA CCG GAG AAA 2159
Lys Asp Ile Ser Leu Asn Leu Glu Val Leu Val Arg Pro Glu Lys
740 745 750

AGG GTG AAG ACG TTG AAA ATC TTG GGA TTG ATG ACA AAA GCA AAT 2204
Arg Val Lys Thr Leu Lys Ile Leu Gly Leu Met Thr Lys Ala Asn
755 760 765

TCG AGA ATG CTG TTA TGT TCA TGG TCT AAT GGT GTC CAT AAG ATG 2249
Ser Arg Met Leu Leu Cys Ser Trp Ser Asn Gly Val His Lys Met
770 775 780

CTT CGG AAG GCT CGG TTC TCT GAC AAA GTA GAT CAG GCT TCT AGT 2294
Leu Arg Lys Ala Arg Phe Ser Asp Lys Val Asp Gln Ala Ser Ser
785 790 795

AAA TAT CCA GCA TTT AGG AGG AGA AAA CTT ATA TAT GTT ATT GCT 2339
Lys Tyr Pro Ala Phe Arg Arg Arg Lys Leu Ile Tyr Val Ile Ala
800 805 810

GTA GAC GGG GAT TAT GAA GAT GGA CTT TTT GAT ATT GTT CGG AGG 2384
Val Asp Gly Asp Tyr Glu Asp Gly Leu Phe Asp Ile Val Arg Arg
815 820 825

62

ATA	TTT	GAT	GCT	GCT	GGC	AAG	GAG	AAG	ATT	GAA	GGT	TCC	ATC	GGG	2429
Ile	Phe	Asp	Ala	Ala	Gly	Lys	Glu	Lys	Ile	Glu	Gly	Ser	Ile	Gly	
					830				835					840	

TTT	ATA	TTG	TCA	ACA	TCC	TAT	TCT	ATG	CCC	GAA	ATT	CAG	AAC	TAT	2474
Phe	Ile	Leu	Ser	Thr	Ser	Tyr	Ser	Met	Pro	Glu	Ile	Gln	Asn	Tyr	
					845				850					855	

TTG	CTA	TCA	AAA	GGC	TTC	AAT	CTT	CAT	GAT	TTT	GAT	GCA	TAT	ATA	2519
Leu	Leu	Ser	Lys	Gly	Phe	Asn	Leu	His	Asp	Phe	Asp	Ala	Tyr	Ile	
					860				865					870	

TGC	AAC	AGT	GGG	AGT	GAG	TTG	TAC	TAT	TCA	TCT	TTG	AAC	TCA	GAG	2564
Cys	Asn	Ser	Gly	Ser	Glu	Leu	Tyr	Tyr	Ser	Ser	Leu	Asn	Ser	Glu	
					875				880					885	

GAG	AGT	AAT	ATT	ATA	GCA	GAT	TCA	GAT	TAC	CAT	TCA	CAC	ATA	GAG	2609
Glu	Ser	Asn	Ile	Ile	Ala	Asp	Ser	Asp	Tyr	His	Ser	His	Ile	Glu	
					890				895					900	

TAC	AGA	TGG	GGT	GGA	GAA	GGC	CTT	AGA	AGG	ACT	TTG	CTT	CGC	TGG	2654
Tyr	Arg	Trp	Gly	Gly	Glu	Gly	Leu	Arg	Arg	Thr	Leu	Leu	Arg	Trp	
					905				910					915	

GCA	GCT	TCC	ATC	ACA	GAA	AAA	AAT	GGT	GAA	AAC	GAA	GAA	CAG	GTT	2699
Ala	Ala	Ser	Ile	Thr	Glu	Lys	Asn	Gly	Glu	Asn	Glu	Glu	Gln	Val	
					920				925					930	

63

ATT ACT GAA GAT GAA GAA GTT TCT ACG GGT TAT TGC TTT GCG TTT 2744
 Ile Thr Glu Asp Glu Glu Val Ser Thr Gly Tyr Cys Phe Ala Phe
 935 940 945

AAA ATA AAG AAC CAA AAT AAG GTT CCC CCT ACG AAG GAG CTC CGC 2789
 Lys Ile Lys Asn Gln Asn Lys Val Pro Pro Thr Lys Glu Leu Arg
 950 955 960

AAG TCA ATG AGG ATT CAA GCT CTT CGT TGC CAT GTG ATT TAC TGT 2834
 Lys Ser Met Arg Ile Gln Ala Leu Arg Cys His Val Ile Tyr Cys
 965 970 975

CAG AAC GGA TCT AAA ATG AAT GTG ATT CCA GTA CTA GCA TCC CGT 2879
 Gln Asn Gly Ser Lys Met Asn Val Ile Pro Val Leu Ala Ser Arg
 980 985 990

TCT CAA GCC CTC AGG TAT CTT TAT GTT CGT TGG GGA GTT GAG TTG 2924
 Ser Gln Ala Leu Arg Tyr Leu Tyr Val Arg Trp Gly Val Glu Leu
 995 1000 1005

TCG AAG ATG GTT GTC TTT GTT GGA GAA TGT GGT GAC ACA GAT TAT 2969
 Ser Lys Met Val Val Phe Val Gly Glu Cys Gly Asp Thr Asp Tyr
 1010 1015 1020

GAA GGC TTG CTT GGC GGG GTC CAT AAA ACC GTA ATA CTG AAG GGA 3014
 Glu Gly Leu Leu Gly Gly Val His Lys Thr Val Ile Leu Lys Gly
 1025 1030 1035

64

GTC TCC AAC ACT GCT TTA AGG TCT CTC CAT GCC AAC AGA AGT TAC 3059
Val Ser Asn Thr Ala Leu Arg Ser Leu His Ala Asn Arg Ser Tyr
1040 1045 1050

CCT CTT TCA CAT GTC GTG TCG CTT GAC AGC CCC AAT ATT GGC GAG 3104
Pro Leu Ser His Val Val Ser Leu Asp Ser Pro Asn Ile Gly Glu
1055 1060 1065

GTG AGC AAA GGG TGC AGC AGC TCC GAG ATC CAG TCC ATC GTC ACA 3149
Val Ser Lys Gly Cys Ser Ser Ser Glu Ile Gln Ser Ile Val Thr
1070 1075 1080

AAA CTC TCC AAA GCT TAA TCAGATATCT GCTGCTTTCT TTTGGGTAAG 3197
Lys Leu Ser Lys Ala End
1085

CAAGGTTTCA TCTTATATGA TTATATCATA AGATACTATA TAAGCACCTT 3247

ATTGGTAAGT CAGTCCCATA ATAATAATGT ACTTCAGAAC CACAATACTT 3297

AAAAGTTGGT TCAGTAGTGA TTAGTCTCAT AATAATCATA TAATTACACA 3347

TCCGCTGTTA ACTAGTGGTA ATATCTAAGC TCAACAATAA AGATGTAAAA 3397

TGCTAGTATG GAAATGAATT GCTAGCTGTT GATCTCTTTC CCTTTATTCT 3447

GTATTATTTTCT TTTCCTCATC TCATGTAAAA ACAATTTTCT GAAGGTGTAC 3497

AGTTTTTTTCC CCTTATATAT CTGTATTATT TCTACTATTT TTTGTTTGTA 3547

AGAATATCCT CTCATCGAGG AGTGATAATT AAATAACCGG CTTGCTAAAT 3597

65

ATAAAGCTTA TTCGAGTTAA AAAAAAAAAA AAAAAAAAAA

3635

4. DNA sequence with the coding region for
sucrose-synthase for the preparation of sugar beet
with modified sucrose concentration, characterised
in that the sequence has the following nucleotide
sequence (Seq. ID No.4):

CT GCA GGA GGG AAA CAA ATT CTT AGC GAT GGC CCG TTT AGC GAA 0044
Ala Gly Gly Lys Gln Ile Leu Ser Asp Gly Pro Phe Ser Glu
5 10

GTT CTT AGG TCT GCT CAG GAA GCA ATA GTT GTT CCT CCC TTT GTT 0089
Val Leu Arg Ser Ala Gln Glu Ala Ile Val Val Pro Pro Phe Val
15 20 25

GCT ATA GCA GTC CGT CCA AGA CCT GGA GTT TGG GAA TAT GTT CGT 0134
Ala Ile Ala Val Arg Pro Arg Pro Gly Val Trp Glu Tyr Val Arg
30 35 40

GTT AAT GTC TCT GAA CTG AAT GTG GAG CAG CTA ACT GTG TCT GAG 0179
Val Asn Val Ser Glu Leu Asn Val Glu Gln Leu Thr Val Ser Glu
45 50 55

TAT CTC CAT TTC AAG GAA GAA CTT GTG GAT GGA AAG GCT GAT GAC 0224
Tyr Leu His Phe Lys Glu Glu Leu Val Asp Gly Lys Ala Asp Asp
60 65 70

CAC TAT GTG CTT GAG CTT GAT TTC GAG CCT TTT AAT GAA TCA GTT 0269
His Tyr Val Leu Glu Leu Asp Phe Glu Pro Phe Asn Glu Ser Val
75 80 85

CCA CGT CCA ACT CGC TCT TCA TCA ATT GGT AAT GGT GTT CAG TTC 0314
Pro Arg Pro Thr Arg Ser Ser Ser Ile Gly Asn Gly Val Gln Phe
90 95 100

66

CTC	AAT	CGG	CAC	CTG	TCA	TCA	AGC	ATG	TTC	TGC	AAC	AAA	GAT	TGC	0359
Leu	Asn	Arg	His	Leu	Ser	Ser	Ser	Met	Phe	Cys	Asn	Lys	Asp	Cys	
105					110					115					

TTG	GAG	CCG	TTA	CTT	GAT	TTT	CTT	AGA	GTG	CAC	AAA	CAT	AAA	GGA	0404
Leu	Glu	Pro	Leu	Leu	Asp	Phe	Leu	Arg	Val	His	Lys	His	Lys	Gly	
120					125					130					

GTT	GTC	ATG	ATG	TTG	AAT	GAT	CGG	ATA	CAG	ACT	ATC	CAG	CGT	CTT	0449
Val	Val	Met	Met	Leu	Asn	Asp	Arg	Ile	Gln	Thr	Ile	Gln	Arg	Leu	
135					140					145					

CAG	TCT	GCA	TTG	TCT	AAA	GCT	GAG	GAT	TAT	CTT	ATC	AAA	CTT	CCA	0494
Gln	Ser	Ala	Leu	Ser	Lys	Ala	Glu	Asp	Tyr	Leu	Ile	Lys	Leu	Pro	
150					155					160					

GCA	GAT	ACA	CCT	TAC	TCT	GAG	TTC	GAA	TTT	GTA	ATC	CAA	GGT	ATG	0539
Ala	Asp	Thr	Pro	Tyr	Ser	Glu	Phe	Glu	Phe	Val	Ile	Gln	Gly	Met	
165					170					175					

GGT	TTT	GAA	AGA	GGC	TGG	GGT	GAT	ACT	GCT	GAA	AGG	GTT	CTA	GAA	0584
Gly	Phe	Glu	Arg	Gly	Trp	Gly	Asp	Thr	Ala	Glu	Arg	Val	Leu	Glu	
180					185					190					

ATG	ATG	CAT	CTA	CTA	CTA	GAT	ATC	CTT	CAG	GCT	CCC	GAT	CCG	TCT	0629
Met	Met	His	Leu	Leu	Leu	Asp	Ile	Leu	Gln	Ala	Pro	Asp	Pro	Ser	
195					200					205					

67

ACA TTA GAG ACA TTT CTG GGA AGA CTT CCC ATG GTG TTT AAT GTG 0674
 Thr Leu Glu Thr Phe Leu Gly Arg Leu Pro Met Val Phe Asn Val
 210 215 220

GTC ATT TTG TCT GTA CAT GGA TAT TTT GGA CAG GCA CAT GTG CTC 0719
 Val Ile Leu Ser Val His Gly Tyr Phe Gly Gln Ala His Val Leu
 225 230 235

GGC TTG CCT GAC ACT GGT GGG CAG ATA GTT TAT ATA CTT GAC CAA 0764
 Gly Leu Pro Asp Thr Gly Gly Gln Ile Val Tyr Ile Leu Asp Gln
 240 245 250

GTG CGG TCT CTG GAA CAT GAA ATG CTC CAA CGA ATA AAG AAG CAA 0809
 Val Arg Ser Leu Glu His Glu Met Leu Gln Arg Ile Lys Lys Gln
 255 260 265

GGA CTA GAT GTG ACT CCT AGA ATT CTT ATC GTG AGT CGG TTG ATT 0854
 Gly Leu Asp Val Thr Pro Arg Ile Leu Ile Val Ser Arg Leu Ile
 270 275 280

CCT GAC GCT AAA GGG ACC ACG TGC AAT CAA CGT ATG GAG AAA GTC 0899
 Pro Asp Ala Lys Gly Thr Thr Cys Asn Gln Arg Met Glu Lys Val
 285 290 295

AGT GGA ACA GAG CAT GCT AGT ATC CTG AGA GTT CCT TTC CGA TCA 0944
 Ser Gly Thr Glu His Ala Ser Ile Leu Arg Val Pro Phe Arg Ser
 300 305 310

68

GAG AAA GGA ATC CTC CGC AAA TGG ATA TCT AGA TTT GAT GTA TGG 0989
 Glu Lys Gly Ile Leu Arg Lys Trp Ile Ser Arg Phe Asp Val Trp
 315 320 325

CCT TAT TTA GAG ACC TTC ACT GAG GAT GCA GCT GGT GAA ATT ATT 1034
 Pro Tyr Leu Glu Thr Phe Thr Glu Asp Ala Ala Gly Glu Ile Ile
 330 335 340

GGC GAG TTG CAG GGT CGT CCA GAT CTG ATA ATT GGC AAC TAC AGC 1079
 Gly Glu Leu Gln Gly Arg Pro Asp Leu Ile Ile Gly Asn Tyr Ser
 345 350 355

GAT GGG AAT ATA GTT GCT TCT TTA TTG TCC CAC AAA ATG GGT GTC 1124
 Asp Gly Asn Ile Val Ala Ser Leu Leu Ser His Lys Met Gly Val
 360 365 370

ACC CAG TGC AAT ATA GCC CAT GCA TTG GAG AAA ACC AAG TAT CCA 1169
 Thr Gln Cys Asn Ile Ala His Ala Leu Glu Lys Thr Lys Tyr Pro
 375 380 385

GAT TCT GAT ATT TAC TGG AAA AGA TTT GAG GAC AAA TAT CAC TTC 1214
 Asp Ser Asp Ile Tyr Trp Lys Arg Phe Glu Asp Lys Tyr His Phe
 390 395 400

TCG TGT CAA TTT TCA GCT GAC TTG ATG GCA ATG AAT CAT GCT GAT 1259
 Ser Cys Gln Phe Ser Ala Asp Leu Met Ala Met Asn His Ala Asp
 405 410 415

TTC ATC ATT ACG AGT ACT TAC CAA GAG ATA GCT GGA ACG AAG AAT 1304
Phe Ile Ile Thr Ser Thr Tyr Gln Glu Ile Ala Gly Thr Lys Asn
420 425 430

CTG TAT CGG GTG GTT CAC GGG ATT GAT GTC TTT GAT CCC AAG TTT 1394
Leu Tyr Arg Val Val His Gly Ile Asp Val Phe Asp Pro Lys Phe
450 455 460

TCA GAG AAG GAT GTC ACC TGT CTC ACT TCA CTT CAT AGA CTT ATA 1484
Ser Glu Lys Asp Val Thr Cys Leu Thr Ser Leu His Arg Leu Ile
480 485 490

GTA TTA GAT GAT ACC TCA AAG CCA ATT ATA TTT TCC ATG GCG AGG 1574
Val Leu Asp Asp Thr Ser Lys Pro Ile Ile Phe Ser Met Ala Arg
510 515 520

70

CTA GAC CGT GTG AAG AAT ATA ACA GGG CTG GTA GAG TGC TAT GGC 1619
Leu Asp Arg Val Lys Asn Ile Thr Gly Leu Val Glu Cys Tyr Gly
525 530 535

AAG AAT GCG AAA CTC AGG GAA CTG GCA AAC CTG GTT GTA GTG GCT 1664
Lys Asn Ala Lys Leu Arg Glu Leu Ala Asn Leu Val Val Val Ala
540 545 550

GGG TAC AAT GAT GTA AAA AAG TCG AAT GAC AGG GAG GAA ATT GCC 1709
Gly Tyr Asn Asp Val Lys Lys Ser Asn Asp Arg Glu Glu Ile Ala
555 560 565

GAA ATC GAG AAG ATG CAC AGG CTT ATA CAG GAG TAT AAT TTA AGA 1754
Glu Ile Glu Lys Met His Arg Leu Ile Gln Glu Tyr Asn Leu Arg
570 575 580

GGA CAA TTT CGC TGG ATT GCT TCT CAA ACA AAT AGA GTA CGA AAT 1799
Gly Gln Phe Arg Trp Ile Ala Ser Gln Thr Asn Arg Val Arg Asn
585 590 595

GGT GAA CTC TAT CGC TAC ATT TGT GAC AAA GGA GGT ATT TTT GCG 1844
Gly Glu Leu Tyr Arg Tyr Ile Cys Asp Lys Gly Gly Ile Phe Ala
600 605 610

CAG CCT GCA TTT TAT GAA GCA TTT GGG CTT ACA GTT GTT GAA GCC 1889
Gln Pro Ala Phe Tyr Glu Ala Phe Gly Leu Thr Val Val Glu Ala
615 620 625

71

ATG ACC TGT GGT CTT CCC ACA TTT GCT ACC TGC CAC GGT GGT CCA 1934
Met Thr Cys Gly Leu Pro Thr Phe Ala Thr Cys His Gly Gly Pro
630 635 640

GCT GAG ATT ATA GAA GAC GGT GTT TCA GGA TTT CAT ATC GAT CCA 1979
Ala Glu Ile Ile Glu Asp Gly Val Ser Gly Phe His Ile Asp Pro
645 650 655

TAT CAT GCT GAT CAG GCA GAA AAA ATG ACT GAA TTC TTT GTC AAG 2024
Tyr His Ala Asp Gln Ala Glu Lys Met Thr Glu Phe Phe Val Lys
660 665 670

TGC AGA GAG GAT CCA AAC TAC TGG ACT AAA ATC TCT GCA GGA GGG 2069
Cys Arg Glu Asp Pro Asn Tyr Trp Thr Lys Ile Ser Ala Gly Gly
675 680 685

TTA CTA AGG ATC AAA GAA AGA TAT ACC TGG CAA AAG TAT TCT GAA 2114
Leu Leu Arg Ile Lys Glu Arg Tyr Thr Trp Gln Lys Tyr Ser Glu
690 695 700

AGG TTA ATG ACA TTG GCA GGG GTG TAT GGT TTC TGG AAA TAT GTC 2159
Arg Leu Met Thr Leu Ala Gly Val Tyr Gly Phe Trp Lys Tyr Val
705 710 715

TCT AAA CTA GAG AGA AGA GAG ACA CGA CGT TAT CTT GAG ATG TTC 2204
Ser Lys Leu Glu Arg Arg Glu Thr Arg Arg Tyr Leu Glu Met Phe
720 725 730

72

TAC ATT TTG AAG TTC CGT GAT CTG GCC AAC TCT GTT CCG CTG GCA 2249
 Tyr Ile Leu Lys Phe Arg Asp Leu Ala Asn Ser Val Pro Leu Ala
 735 740 745

ACA GAT GAA GAG CCT TCT ACT ACT GAT GCA GTT GCG ACA TTC CGT 2294
 Thr Asp Glu Glu Pro Ser Thr Thr Asp Ala Val Ala Thr Phe Arg
 750 755 760

GGA CCT TGA ACGCTGCTGC TTACTGAGGT TCCAAGTTGT GTATATATTA 2343
 Gly Pro End

CTGTGAAAGG AATAAGTGTA GCTACACAAA AGGTTCTCAA CTATTAGTAT 2393

CTTCTCTGTG TAAATAACGA GAGTGAAAAA TGTAATATTG TTGATGTCTT 2443

GAAAACTGAG TTTGCTTTGT TTATTTTAA GTGTATGACA ATATGTATCA 2493

TATAACGGAT TCTTCAGTGA TCATATCAAA AACTACTGAC CATCGAAGTT 2543

AATGAAAATC GACAGCAACA 2563

5. Derivatives of DNA sequences according to any one of
 claims 1 to 4 characterised in that these
 derivatives are obtained by exchange of single bases
 5 or by targeted or non-targeted mutagenesis.

6. Plasmids containing
 a) a suitable promoter which ensures that the
 coding sequence meets a suitable time point or
 10 in the specified developments in the transgenic
 plant or in determined genes of transgenic
 plants;

- b) at least one coding sequence according to claims 1 to 5, that
- i) is so coupled to the promoter that the formation of an RNA is allowed which is into a protein, whereby the protein demonstrates an enzymatic activity which leads to a modification of the sucrose concentration in the plant, or
 - ii) which is so coupled to the promoter that the coding part is read, which leads to the formation of a so-called anti-sense RNA which under-expresses the formation of the protein coded from an endogenous gene in the plant, that is involved in the sucrose biosynthesis; and
- c) A non-coding termination sequence that contains the signal for the termination and poly-adenylation of the transcript.
7. Use of the DNA-sequences according to any one of claims 1 to 6 for the preparation of sugar beet with changed sucrose concentration.
8. Use of the plasmids according to any one of claims 1-4 for the preparation of derivatives by targeted or non-targeted mutagenesis
9. Sugar beet containing a DNA sequence according to any one of claims 1-5.

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(21) International Application Number: PCT/EP94/01671 (22) International Filing Date: 20 May 1994 (20.05.94) (30) Priority Data: P 43 17 596.1 24 May 1993 (24.05.93) DE (71) Applicant (for all designated States except US): HOECHST SCHERING AGREVO GMBH [DE/DE]; Gerichtstrasse 27, D-13347 Berlin (DE). (72) Inventors; and (75) Inventors/Applicants (for US only): HESSE, Holger [DE/DE]; Brundhildstrasse 9, D-10829 Berlin (DE). MÜLLER- RÖBER, Bernd [DE/DE]; Siemensstrasse 71, D-12247 Berlin (DE).		(81) Designated States: CA, HU, JP, RU, UA, US, European patent (AT, BE, CH, DE, DK, ES, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE). Published <i>With international search report.</i> <i>Before the expiration of the time limit for amending the claims and to be republished in the event of the receipt of amendments.</i> (88) Date of publication of the international search report: 26 January 1995 (26.01.95)
(54) Title: DNA SEQUENCES AND PLASMIDS FOR THE PREPARATION OF SUGAR BEET WITH CHANGED SUCROSE CONCENTRATION		
(57) Abstract DNA sequences and plasmids are described, that by integration in a plant genome of sugar beet change the sucrose concentration, as well as transgenic plants that by introduction of the DNA sequences of the invention causes changes in sugar concentration.		

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A. CLASSIFICATION OF SUBJECT MATTER
IPC 5 C12N15/82 C12N15/54 A01H5/00

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B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC 5 C12N A01H

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
X	EP,A,0 455 316 (INSTITUT FUR GENBIOLOGISCHE FORSCHUNG) 6 November 1991	5
A	see the whole document ---	1,2
X	WO,A,91 19806 (MONSANTO) 26 December 1991	5
	see page 16 - page 26; figures 5,6 ---	
A	WO,A,93 09237 (SANDOZ) 13 May 1993	1,2
	see the whole document ---	
X	EMBL SEQUENCE DATABASE ACC. NO. S54379 Sucrose phosphate synthase release 35; 31-MAR-1993 ---	5
X	EP,A,0 466 995 (ROUSSEL-UCLAF) 22 January 1992	5
A	see the whole document ---	3
	--- -/--	

☒ Further documents are listed in the continuation of box C.

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C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT		
Category *	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WO,A,92 16631 (ROUSSEL-UCLAF) 1 October 1992 see the whole document ---	3
A	EP,A,0 530 978 (ADVANCED TECHNOLOGIES) 10 March 1992 see the whole document ---	3
X	GENE, vol.60, 1987, AMSTERDAM NL pages 47 - 56 SALANOUBAT, M., ET AL. 'Molecular cloning and sequencing of sucrose synthase cDNA from potato (<i>Solanum tuberosum</i> L.): preliminary characterization of sucrose synthase mRNA distribution' see figure 4 ---	5
A	BIOLOGICAL ABSTRACTS, vol. 95 Philadelphia, PA, US; abstract no. 79002, SAKALO, V.D., ET AL. 'Characterization of molecular forms of sucrose synthase from beet roots' see abstract & FIZIOL RAST (MOSC), vol.39, no.2, 1992 pages 290 - 299 ---	4
A	DATABASE BIOSIS BIOSCIENCES INFORMATION SERVICE, PHILADELPHIA, PA, US DN BR35:113623 MURAKAMI, T. 'Histochemical localization of UDPG ADPG-pyrophosphorylase and phosphorylase in young petiole of sugar beet <i>Beta-vulgaris</i> L.' see abstract & J. HISTOCHEM. CYTOCHEM., vol.36, no.7A, 1988 page 927 ---	1,2
A	EP,A,0 438 904 (ADVANCED TECHNOLOGIES) 31 July 1991 see the whole document --- -/--	1-4

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	<p>BIOLOGICAL ABSTRACTS, vol. 85 Philadelphia, PA, US; abstract no. 72586, FIEUW, S., ET AL. 'Sucrose synthase and sucrose phosphate synthase in sugar beet plants Beta-vulgaris -ssp-altissima' see abstract & J. PLANT PHYSIOL., vol.131, no.1-2, 1987 pages 153 - 162</p> <p>-----</p>	3,4

Form PCT/ISA/210 (continuation of second sheet) (July 1992)

Patent document cited in search report	Publication date	Patent family member(s)		Publication date
EP-A-0455316	06-11-91	DE-A-	4013144	24-10-91
		AU-B-	645302	13-01-94
WO-A-9119806	26-12-91	AU-B-	644203	02-12-93
		AU-A-	8220291	07-01-92
		EP-A-	0536293	14-04-93
WO-A-9309237	13-05-93	AU-A-	2920392	07-06-93
		EP-A-	0611395	24-08-94
EP-A-0466995	22-01-92	AU-B-	653165	22-09-94
		AU-A-	8394591	18-02-92
		WO-A-	9201782	06-02-92
		JP-T-	5502169	22-04-93
WO-A-9216631	01-10-92	AU-A-	8765591	21-10-92
		EP-A-	0533849	31-03-93
EP-A-0530978	10-03-93	NONE		
EP-A-0438904	31-07-91	AU-A-	6836590	04-07-91
		JP-A-	4341126	27-11-92